A DISSERTATION ON

"ESTIMATION OF SERUM VITAMIN 25 (OH) D LEVELS IN CHRONIC LIVER DISEASE PATIENTS AND ITS CLINICAL CORRELATION, A CROSS SECTIONAL STUDY AT A TERTIARY CARE , GOVERNMENT ROYAPETTAH HOSPITAL/ GOVERNMENT KILPAUK MEDICAL COLLEGE IN CHENNAI "

Dissertation Submitted to
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KILPAUK MEDICAL COLLEGE, CHENNAI
THE TAMIL NADU DR.M.G.R.MEDICAL UNIVERSITY
TAMILNADU, INDIA
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CERTIFICATE

This is to certify that this dissertation entitled **"ESTIMATION OF SERUM VITAMIN 25(OH)D LEVEL IN CHRONIC LIVER DISEASE PATIENTS AND ITS CLINICAL CORRELATION,A CROSS SECTIONAL STUDY AT A TERTIARY CARE GOVERNMENT ROYAPETTAH HOSPITAL/ GOVERNMENT KILPAUK MEDICAL COLLEGE IN CHENNAI"** submitted by Dr.B.AMBIKA to the faculty of General Medicine, The Tamil Nadu Dr.M.G.R Medical University, Chennai in partial fulfillment of the requirement for the award of M.D degree Branch I (General Medicine) is a bonafide research work carried out by her under my direct supervision and guidance.

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DECLARATION

I, Dr.B.AMBIKA , solemnly declare that Dissertation titled **"ESTIMATION OF SERUM VITAMIN 25(OH)D LEVEL IN CHRONIC LIVER DISEASE PATIENTS AND ITS CLINICAL CORRELATION,A CROSS SECTIONAL STUDY AT A TERTIARY CARE GOVERNMENT ROYAPETTAH HOSPITAL/ GOVERNMENT KILPAUK MEDICAL COLLEGE IN CHENNAI "** is a bonafide work done by me at Government Royapettah Hospital/ Kilpauk Medical College, Chennai, during April 2017 to September 2017 under the guidance and supervision of Prof. Dr.Venkateswarlu, M.D.DCH, Professor of Medicine, Government Royapettah Hospital,, Chennai. I also declare that this bonafide work or a part of this work was not submitted by me or any other for award degree or diploma to any other university, board either in India or abroad.

This dissertation is submitted to the Tamilnadu DR. M.G.R Medical University, towards partial fulfillment of requirement for the award of M.D. Degree (Branch – I) in General Medicine.

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Date: **Signature of the Candidate**

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ABBREVIATIONS

NAFLD	-	Non alcoholic fatty liver
CYP	-	Cytochrome P
MHC	-	Major histo compatibility
PTH	-	Parathyroid hormone
IFN	-	Interferon
TLR	-	Toll like receptor
LPS	-	Lipo polysaccharide
HCV	-	Hepatitis C virus
SVR	-	Sustained viral response
Th	-	T Helper
IL	-	Interleukin
NK	-	Natural killer cell
DHCR	-	Dehydro cholesterol reductase
VDR	-	Vitamin D receptor
DBP	-	Vitamin D Binding protein
TGF	-	Tranforming growth factor
RBV	-	Ribavirin
NASH	-	Non alcoholic steato hepatitis

IR	-	Insulin resistance
FGF	-	Fibroplast growth factor
TIMP	-	Tissue inhibitor metalloproteinase
MELD	-	Model for end stage liver disease
TNF	-	Tumour necrosing factor

INSTITUTIONAL ETHICS COMMITTEE
GOVT. KILPAUK MEDICAL COLLEGE,
CHENNAI-10
Protocol ID. No.02/2017 Meeting held on 17.04.2017

The Institutional Ethical Committee of Govt. Kilpauk Medical College, Chennai reviewed and discussed the application for approval "**Estimation of serum vitamin 25(OH)D level in chronic liver disease patients and its clinical correlation, a cross sectional study at a tertiary care hospital in chennai"** submitted by Dr.B.Ambika, M.D. (General Medicine), PG Student, GKMC, Chennai-10

The Proposal is APPROVED

The Institutional Ethical Committee expects to be informed about the progress of the study any Adverse Drug Reaction Occurring in the Course of the study any change in the protocol and patient information /informed consent and asks to be provided a copy of the final report.

28. 4. 2017.
DEAN
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26/4/17

My Comp<E Colon< ME 1 Sec> Ethical Committee


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INTRODUCTION

INTRODUCTION

Vitamin D is a steroid hormone (secosteriod-where one of the bond in the rings is broken). It has multiple effects and uses in the human body. Vitamin D has anti fibrotic effect and anti-inflammatory effect other than the effects on calcium and skeletal metabolism . This anti-inflammatory and antifibrotic property of vitamin D is implicated in the causation ,disease progression of chronic liver disease. There are many research studies stating that supplementing vitamin D will decrease the severity of chronic liver disease.

The regulation of both innate and acquired immune systems is modulated by Vitamin D. Though synthesised mainly in the liver ,the extra skeletal benefits of vitamin D such as cellular proliferation , cell differentiation and immunomodulation is the one quoted in many studies for its pathogenesis altering effect of chronic liver disease . Vitamin D improves sustained viral response rate in chronic hepatitis C and decreases insulin resistance caused by its deficiency .Insulin resistance is related to the worsening of NAFLD.

In post transplant patients cell rejection is prevented by giving Vitamin D supplements. Vitamin D also has antifibrotic effect against liver injury caused by thiacetamide.

In patient with chronic liver disease 93% have Vitamin D insufficiency whereas one third of these show severe deficiency [1,2].The outcome of chronic liver diseases with the levels of vitamin D will be studied in this dissertation.

REVIEW OF LITERATURE

REVIEW OF LITERATURE

Chronic liver disease , the major cause of morbidity and mortality all over the world. There are various causes for chronic liver diseases like alcohol ,hepatitis, NAFLD ,autoimmune, cryptogenic.etc..

The association of severity for each of the causes will be discussed separately and there are various studies reviewing the significance of vitamin D level and its clinical significance, knowing about the vitamin D metabolism and its effects will give us an idea about the pathogenesis of how Vitamin D deficiency alters the severity of any chronic liver diseases.

VITAMIN D SYNTHESIS

Vitamin D is synthesised in the skin due to sunlight exposure. There involves three steps .Normally vitamin D is synthesised from epidermal cells. The ultraviolet radiation from the sun changes the metabolite of cholesterol present in the skin i.e. 7-dehydroxy cholesterol to cholecalciferol. This vitamin D is stored in the adipocytes (fat cells).Vitamin D from food sources also will be stored in the adipocytes after getting absorped from the intestine[3,4].Some of the stored Vitamin D is 25 hydroxylated by the hepatic enzymes cytochrome P450 isoforms such as CYP2R1,CYP27A1 and forms calcidiol(25 OH D).Melanin decreases the skin synthesis of vitamin D and the usage of sunscreens also favours non penetration of UV radiation into the skin resulting in decrease of the overall synthesis of vitamin D

from skin exposure. Vitamin D from dietary sources and animal sources after absorption binds to vitamin D binding protein and will be subjected to 25 hydroxylation as mentioned before. This enzyme is not a regulated one. Its metabolite 25(OH) vitamin D binds to the alpha globulin vitamin D binding protein. It is the reliable reflector of vitamin D levels in the body for both the circulation and storage forms.

88% of the 25(OH)D will be in this bound state. 0.03% is in the free form,remaining will be bound to the albumin .(t ½) Half life of 25 (OH) vitamin D is around 2-3 weeks. Vitamin D binding protein has also other functions apart from storing 25(OH) D such as anti inflammatory and immuno modulation.[5,6]

The half life of 25 (OH) D is dependent on the Vitamin D.It is dependent on the vitamin D binding protein levels ,which is evident in nephrotic syndrome patients where increased urinary excretion of protein in urine resulting in decreased vitamin D binding protein levels.

The second step involves the production of mature hormone which occurs in the organ kidney. Kidney is one of the vital organ involved in the metabolism of vitamin D and calcium, mainly the bone metabolism.

1-alpha hydroxylase(CYP27B1) in the proximal convoluted lobules of the kidney converts the calcidiol 25(OH) D to 1-alpha 25 (OH)2 D (1-alpha 25 dihydroxy vitamin D) calcitriol which is the most biologically active form of Vitamin D.[7]

These water soluble metabolites are secreted into bile acids and reabsorbed back through the enterohepatic circulation.CYP24A1 is the isoform and any

mutation in this gene (cytochrome P450) causes infantile hypercalcemia .Impairment of the enterohepatic recirculation due to terminal ileal diseases causes increased rate of loss of vitamin D metabolites.

Inactivation of vitamin D is by adding one more hydroxyl group by the enzyme 24 hydroxylase ,this is formed mostly in intestine and kidney.1-alpha 25(OH)2D(active form) is having half life of four hours . It is also bound to the vitamin D binding protein in the circulation. This modulates the expressions of 200 genes associated with angiogenesis, apoptosis, proliferation, differentiation and immuno modulation[8,9]. These effects are mediated by the binding of vitamin D to its receptor (VDR) present in liver , pancreatic islets, gastro intestinal tract, immune system,kidney,muscle cells and osteoblasts. That is how vitamin D deficiency has been implicated in cancer, diabetes mellitus, cardiovascular and auto immune diseases.[10]

Calcitriol will also have autocrine function which means they are synthesised by the macrophages, dendritic cells, T and B lymphocytes with the help of CYP27A 1 or CYP27B1 enzymes present in those cells. Innate immune system is improved by the vitamin D whereas adaptive response is inhibited because of its self regulatory effect .It causes synthesis of peptides like cathelicidin [11,12] and beta defensin which are antibiotic substances and also decrease the expression of class II MHC molecules.Fast non genomic responses are induced by the activation of cellular signaling pathways.

REGULATION OF VITAMIN D SYNTHESIS

Vitamin D synthesis is regulated by the following steps

I . Sunlight destroys the excess of Viamin D.[13]

II . 25 Hydroxylation of vitamin D is not regulated so when the intake of vitamin D is more the levels of 25 (OH) D will also rise .So plasma levels of 25(OH) D is used to regulate vitamin D levels.

III. But the enzyme in kidney 1-alpha hydroxylase is a regulated one.

The factors affecting 1-alpha hydroxylase includes serum calcium, phosphate, parathyroid hormones(PTH),fibroblast growth factor 23(FGF23).An increase in serum calcium level suppresses 1-alpha hydroxylase levels either directly or indirectly decreasing the parathyroid hormone levels[14]. An increase in plasma phosphate also suppresses the 1 alpha hydroxylase activity.There is an increase in FGF23 with elevated levels of serum phosphate . FGF23 inhibits 1-alpha 25(OH)2 D production cytokines and growth factors affects the metabolism of 1-alpha 25(OH)2 D[15].

IV. Vitamin D binding protein levels are directly proportional to the active vitamin D levels. It prevents interaction. It also prevents catabolism and excretion of the Vitamin D hormone. Despite the decreased levels of vitamin D binding protein in liver disease, nephrotic syndrome and malnutrition , the levels of 1-alpha 25 (OH)2 D remains the same.

FIGURE-1-VITAMIN D SYNTHESIS

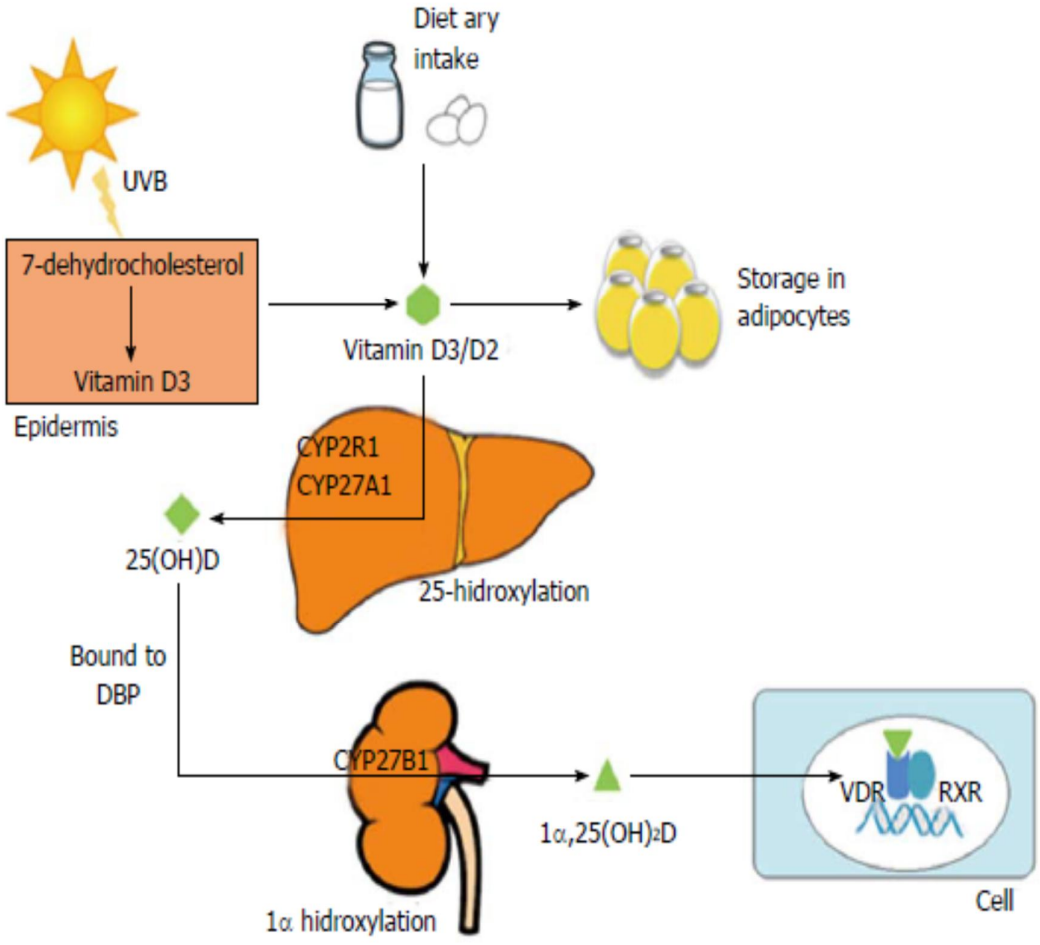
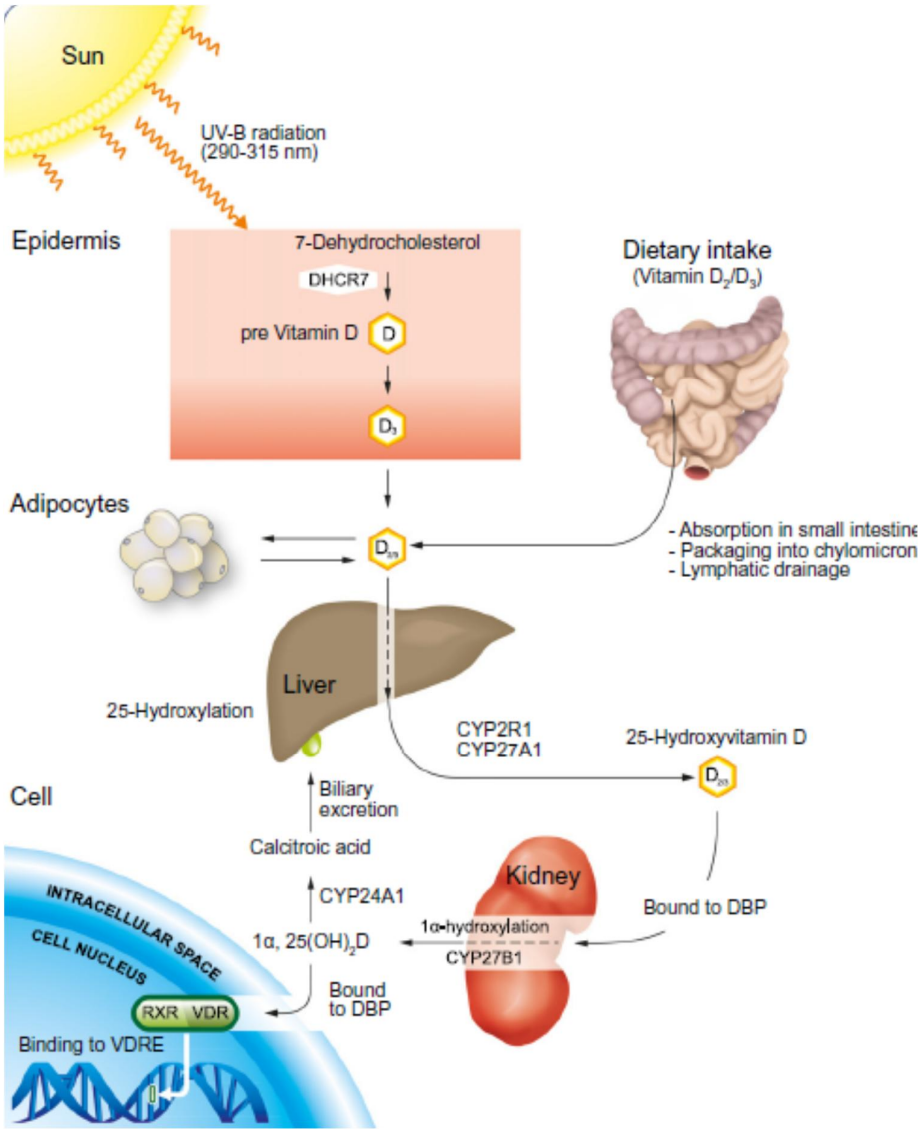


FIGURE-2-VITAMIN D SYNTHESIS



In the macrophages the cytokines like IFN gamma and TLR ligands such as LPS (lipopolysaccharides) will modulate the expression of CYP27B1 and thereby altering the production of 1-alpha 25(OH)2 D

V. CYP24A1(25(OH) VITD-24-Hydroxylase) converts 1-alpha 25 (OH)2 D to calcitroic acid(inactive form).The activity of this enzyme depends on the level of phosphate and parathyroid hormones.

VI . Self regulation effect of 1-alpha 25 (OH)2 D by increasing the expression of CYP24A1 and inhibiting CYP27B1 expression will result in a negative feedback pathway. Thus intake of vitamin D through diet or sunlight is one of the many variables in regulating vitamin D apart from the variables like vitamin D binding protein levels, autocrine effect of 1-alpha 25(OH)2 D ,vitamin D receptor expression .Thus implicating the multi factorial regulation of vitamin D metabolism.

FUNCTIONS OF VITAMIN D

The various effects of vitamin D mainly are hepatic and enterohepatic. Considering the hepatic uses, it helps in preventing liver fibrosis when vitamin D supplementation is given in post transplant patients and also the risk of acute rejection is decreased[16] .

In NAFLD patients impairment of liver function is observed in preclinical studies. Main function is the improvement of sustained viral responsive rate(SVR) in HCV affected patients.SVR in HCV is associated with vitamin D binding protein levels. .Extrahepatic function includes mainly immunological modulation[17-19]. In the acquired immune response it shifts the balance to TH12 phenotype by inhibiting the Th1 cells proliferation[20]. Treg cells production are increased and activates naive T Cells and inhibits Th 17 Cells.[21]

Vitamin D supplementation decreases the risk of developing type 1 diabetes in children .In the innate immune system it increases secretion of IL 10 and decreases secretion of IL 12 , increases NK cell cytotoxicity , activates macrophages TLR response to TB infection[22] .Pancreatic or adipocyte functions includes activation of insulin gene transcription resulting in lower prevalence rate of metabolic syndrome in individuals with normal Vitamin D status[23].

BMI is also inversely related to vitamin D levels , higher 25 OH D levels is associated with lesser incidence of colorectal adenoma and Non Hodgkins lymphoma.

Vitamin D deficiency causes more severe steatosis, inflammation and fibrosis. In NAFLD patients the pathogenesis implicated are the increased ILR 2 ,TR 4 and TR 9 expression in liver .In HCV affected individuals poor response to interferon therapy is observed. The activity of the enzyme 25 hydroxylase CYP27A1 in liver is down regulated, and fibrosis progression will become rapid.

It is also the cause of presence of insulin resistance and hyperglycemia due to impaired pancreatic B cell function.

There is also evidence present for the increased risk of breast ,prostate and colonic cancer.

Autoimmune disorder such as multiple sclerosis also increases with its deficiency and the risk for mycobacterium tuberculosis infection also increases. Overall mortality will be decreased by 7% .

It enhances phosphate and calcium absorption in the intestine and promotes osteoclast maturation and decreases parathyroid hormones secretion (PTH).

SOURCES OF VITAMIN D

Vitamin D 2 (Ergocalciferol) is rich in oral supplements, artificially fortified foods e.g. milk, cereals, UV irradiated mushrooms whereas vitamin D3 (Cholecalciferol) is rich in UV B light (290-315nm) ,fishes like salmon,tuna, sardines,,cod liver oil,eggs ,oral supplements ,artificially fortified foods like milk,cereals,bread,margarine .

TABLE-1-SOURCES OF VITAMIN D

Vitamin D2 (ergocalciferol):
UV-irradiated mushrooms
Oral supplements
Artificially fortified foods (e.g. milk, cereals, bread, margarine)
Vitamin D3 (cholecalciferol):
Ultraviolet B light (290-315 nm): the major source of vitamin D3
Fatty fish:
Salmon
Mackerel
Tuna
Sardines
Eel
Cod liver oil
Eggs
Oral supplements
Artificially fortified foods (e.g. milk, cereals, bread, margarine)

CHRONIC LIVER DISEASE AND VITAMIN D

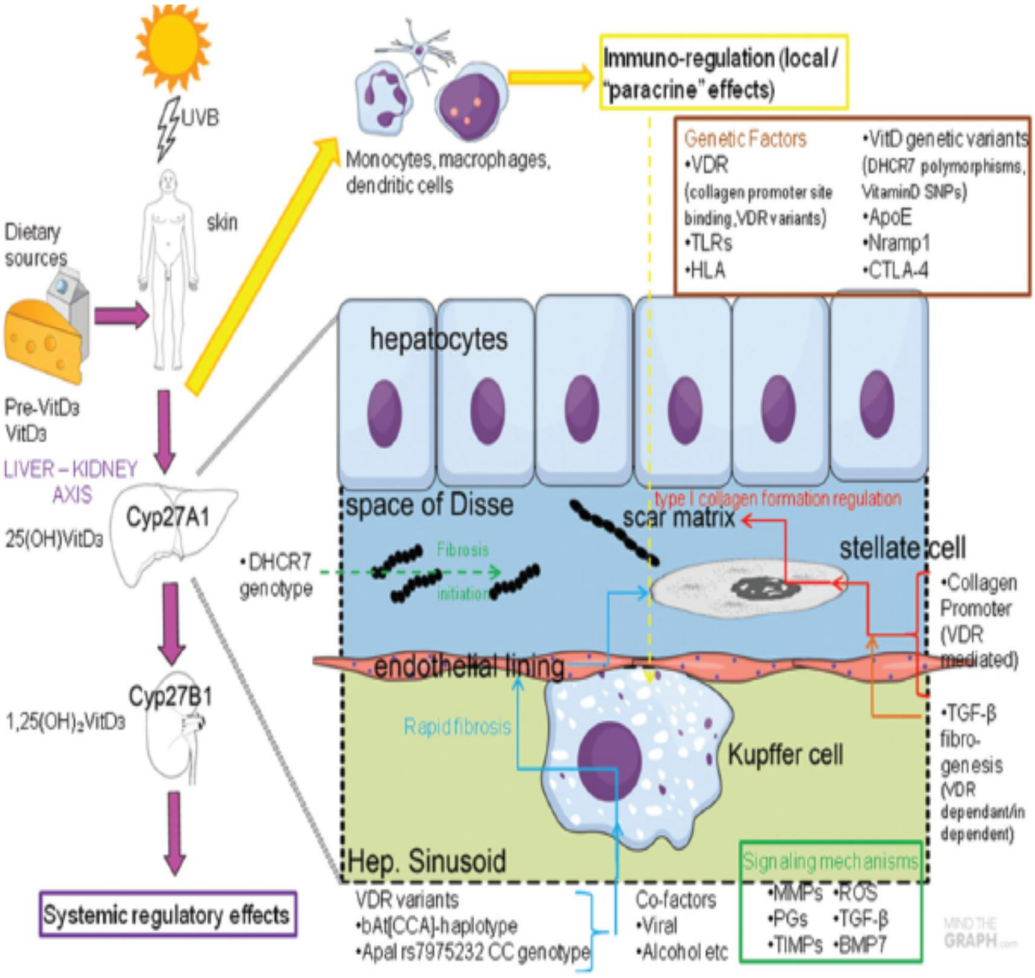
In all the chronic diseases like type 2 diabetes mellitus ,several type of cancers and cardiovascular infectious disease and autoimmune disorders,vitamin D plays an important role in reducing the overall risk of these diseases.

Vitamin D deficiency is more common among the chronic liver disease patients. Patients with milder forms of liver disease are also affected though only cirrhosis causes severe deficiency.

The effects of 1-alpha 25(OH)2 D by its autocrine and paracrine actions results in the proliferation of cells and apoptosis, insulin & renin secretion ,interleukin and bactericidal protein production which can be attributed to the pathogenesis of chronic liver disease since the liver is an important organ in the metabolism of vitaminD. Whether vitamin D deficiency causes liver dysfunction or its deficiency is due to liver disease should be answered as several studies in the population observed that low levels of 25 (OH) D increases the risk of mortality from all causes.

So patients with chronic liver disease of different etiologies vitamin D deficiency causes increase in mortality and morbidity by causing bacterial infection ,fibrosis severity and portal hypertension complication.[24,25]

FIGURE-3-VITAMIN D AND LIVER FIBROSIS



Vitamin D deficiency in chronic liver disease occurs due to many causes apart from cirrhosis like inadequate sun exposure ,steroid use ,inadequate food intake, jaundice ,decreased vitamin D absorption due to intestinal edema because of portal hypertension or due to cholestasis causing bile salt disruption .In severe cirrhosis vitamin D hydroxylation is decreased resulting in decreased albumin and vitamin D binding protein(DBP) production. Synthetic function of liver is affected only in the severely affected patients. Vitamin D Deficiency is also prevalent in non cirrhotic patients.

After vitamin D supplementation the level of 25(OH) D returns to normal levels indicating preserved synthetic function of liver though vitamin D binding protein levels are decreased .But vitamin D requires 5% of vitamin D binding protein binding sites again proving that liver dysfunction should be severe enough to cause decreased vitamin D binding protein levels and vitamin D levels[26].

There is an increasing evidence for involvement of vitamin D in decreasing the inflammation and fibrosis though all the relations between vitamin D and liver disease is not enough to find a causative effect between the severity of chronic liver disease and Vitamin D deficiency.

The systematic and umbrella review says that there exists no doubt about on any causal association between extra skeletal effects and Vitamin D deficiency. Vitamin D deficiency is generally a marker of illness than related to the pathogenesis of these diseases. But there is a relationship between vitamin D and hepatic

physiopathology due to signaling disruptions in the non parenchymal liver cells or extrahepatic cells.

Almost 90% of the macrophages are in liver therefore inflammatory disease of liver will affect the production of active vitamin D. The expression of CYP27B1 is increased resulting in more production of 1-alpha 25 (OH)2 D by the proinflammatory response. Vitamin D receptor is expressed in biliary epithelial cells, non parenchymal cells and macrophages[27,28] .One gets activated cathelicidin which is an anti endoxin inhibits the biliary acid synthesis and protects the hepatocytes from this acids. Genetic factors also contributes to variation in vitamin D Levels .

Serum levels of 25(OH)D and its efficacy have been strongly associated with many simple nucleotide polymorphisms of genes regulating the metabolism of vitamin D receptor and vitamin D like receptor DHCR 7 (which encodes the 7 dehydro cholesterol reductase enzyme) and CYP2R1,CYP24A1,GC (encodes vitamin D binding protein). [29-32]

A study involving black Americans and whites says that black Americans had low levels of vitamin D and vitamin D binding protein ,thus substantiating the common genetic polymorphisms is prevalent in these population and leading on to the racial difference.[33] Primary biliary cirrhosis, auto immune hepatitis, hepatocellular cancer have been associated with genetic polymorphism of vitamin D receptor gene leading onto the severity of chronic liver disease.[34-39]

The vitamin D supplements are currently given for decreasing the skeletal abnormalities due to vitamin D deficiency. The data available says vitamin D supplements are useful in reducing the overall morbidity and mortality .

At least 30 ng/ml will reduce the risk of 25 (OH) D in many studies .In our another study where meta analysis included 73 cohort studies and 22 controlled and randomized studies show vitamin D supplements significantly reduce mortality from any cause in adults. Few prospective studies gives contradictory results regarding the effects of supplementation of vitamin D because of genetic polymorphisms, defective study designs, quantity of vitamin D administration and the pre or post treatment measurements used .

The new recommendation of 25 (OH)D levels is around 20ng/ml which is sufficient to meet the needs .[40] But ESCPG (endocrine society clinical practice guideline) says need may be higher for high risk patients so the level above 30 ng/ml will reduce the risk in these patients.[41] For cholestatic liver disease patients vitamin D supplements of 400-800 U/Day along with calcium (1000-1200 mg/day) is suggested by the ESCPG. [42]

Though vitamin D is deficient in liver disease patients the parathyroid hormone and calcium levels are not affected. This was confirmed in all these patients. Free vitamin D levels is also same as that of the healthy individuals.

So calcium homeostasis is maintained without causing secondary hyperparathyroidism. That is the reason why both free and total 25(OH) D should be measured to detect vitamin D status in chronic liver disease cases.

The decreased bone density in this patients are due to steroid use ,elevated bilirubin levels, hypogonadism, proinflammatory cytokines apart from vitamin D deficiency.

ROLE OF VITAMIN D FUNCTIONS IN LIVER DISEASE

Vitamin D effect on immune system and anti fibrotic effect is implicated in the pathophysiology of chronic liver disease apart from its other uses like skeletal ,CVS,CNS and cellular proliferation and differentiation. In vitro and in vivo mouse models with fibrosis of liver have shown that vitamin D has the antifibrotic property by affecting the process of liver fibrosis in many stages like suppressing the hepatic stellate cell activation and proliferation, inhibiting the injury trigger, reducing the accumulation of extracellular matrix and inhibition of degradation of tissue inhibitor collagen matrix metalloproteinases (TIMPS). [43,44]

Ding et al showed VDR/SMAD geneomic circuit regulating the hepatic fibrogenesis and implicated that VDR ligands in therapy for liver fibrosis because of its wound healing response. Active metabolite of vitamin D 1-alpha 25(OH)2 D prevents liver fibrosis in vivo mice model. But once established cirrhosis cannot be reverted back in animal models.

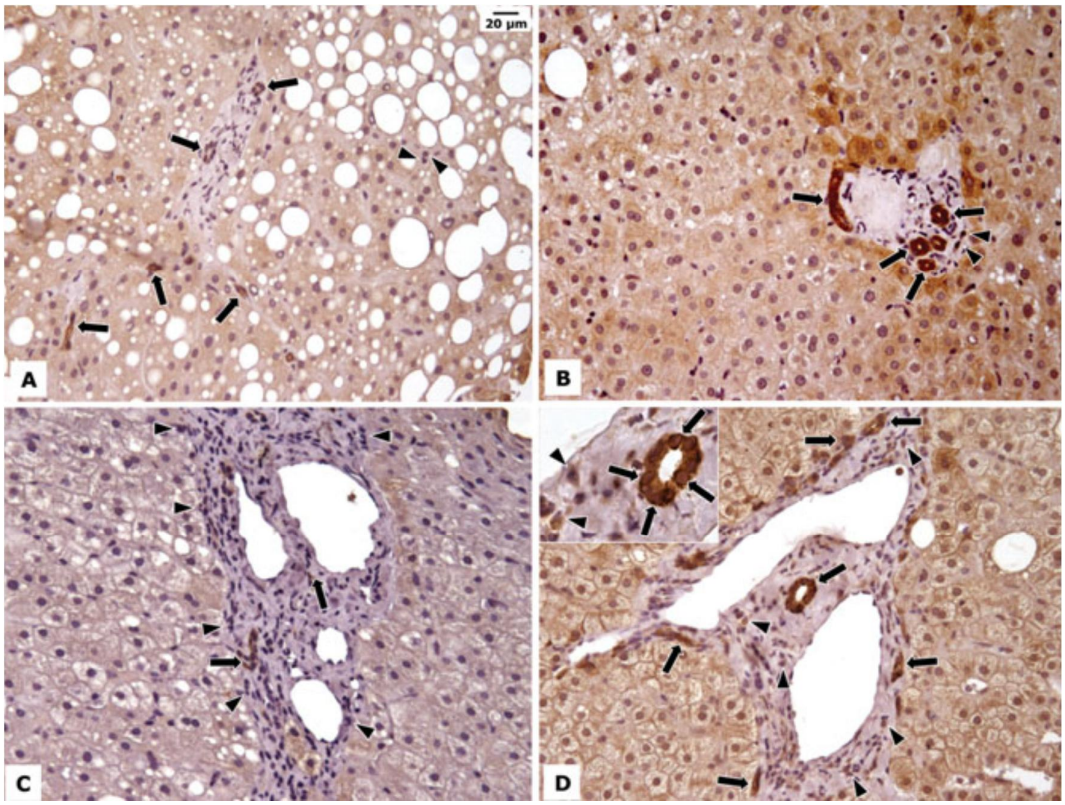


FIGURE-4- -Immunohistochemistry for VDR in NASH and HCV-related CHC. VDR positivity involved few biliary epithelial cells (arrows) and inflammatory cells (arrowheads) in cases with higher NAS in NASH. (A) In cases with severe fibrosis in HCV-related CHC. (B) In which hepatocytes, especially periportal, were also VDRþ and in cases with mild to moderate fibrosis in HCV-related CHC. (C). An increased number of VDRþ biliary epithelial cells (arrows) and inflammatory cells (arrowheads) were observed in cases with lower NAS in NASH. (D) VDR positivity involved the nucleus and cytoplasm (D, inset [high-power field]). Original magnification _200; high-power field magnification _400.

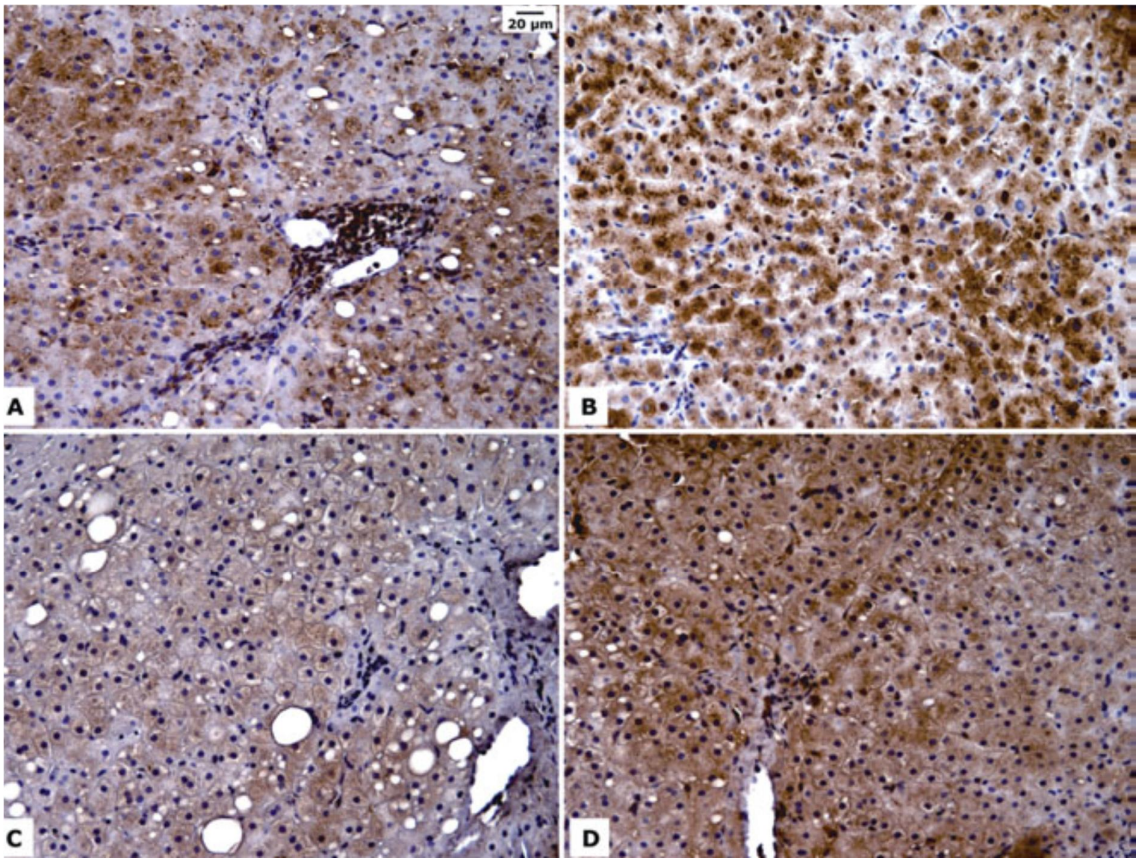


FIGURE-5- Immunohistochemistry for CYP27A1 and CYP2R1 in liver samples. Representative photomicrographs show CYP27A1 (A,B) and CYP2R1 (C,D) expression in the liver. A mild (A,C) and intense (B,D) immunohistochemical score for CYP27A1 and CYP2R1 are presented. Original magnification _200.

Vitamin D regulates the expression of TLR and intestinal permeability . It causes intestinal epithelial cell differentiation and improves cell binding and decrease the bacterial products in the liver . Several studies says regulation of innate and adaptive immune system by vitamin D has got beneficial effects in liver diseases.

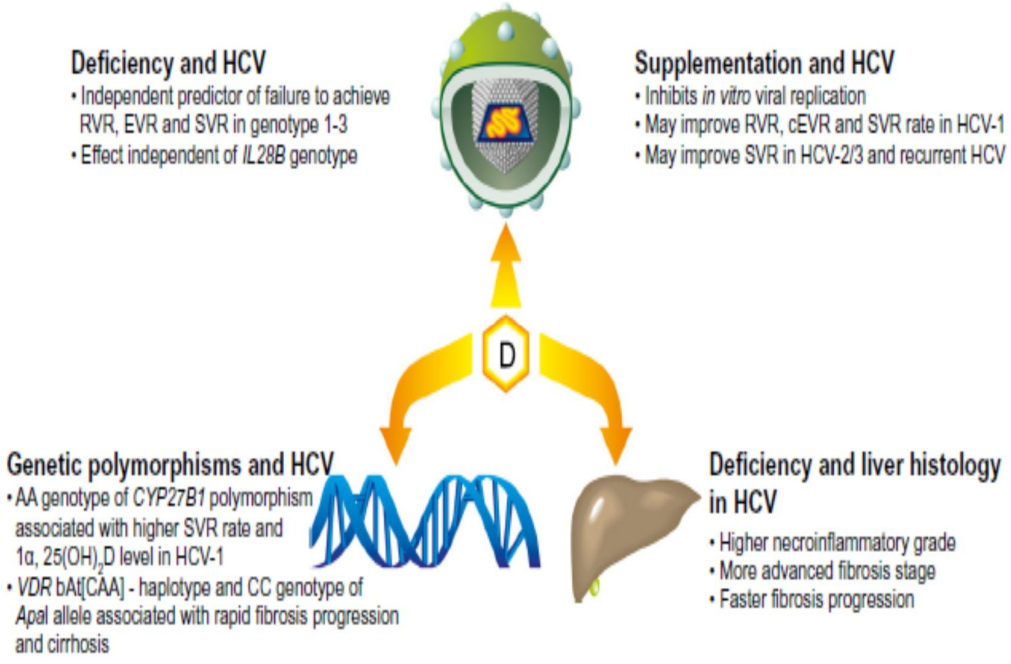
Vitamin D increases the innate immunity by causing chemotaxis and macrophage activation by the release of antibacterial proteins like cathelicidin and beta defensin. There is an inverse relationship between vitamin D levels and the expression of TLR2, TLR4, TLR9 in monocytes.[45,46])These TLRs cause inflammation and fibrosis. After high fat diet and alcohol drinking , structural changes in the mucosa of intestine like loss of epithelial attachment, defects of mucosal immune system, vascular congestion alters the mucosal permeability and increases the intestinal bacterial translocation and once lipopolysaccharides(LPS) enters the blood stream and triggers TLR4 response in the hepatocytes, biliary cells, dendritic cells causing fibrogenesis leading on to liver damage.

Adaptive immunity is related to the association of autoimmune disease with the liver damage to the vitamin D deficiency. Vitamin D decreases the class II HLA expressions and modulates T cell response .The development of Th 17 cells by inhibition of IL-6 IL-23 which are produced by the dentritic cells due to 1- alpha 25 (OH) 2 D and also causes differentiation and proliferation of regulatory T cells which produces cytokines IL- 10 and TGF B .Vitamin D dependent activation of naive T cells also occur. The polarisation of Th Cells is shifted towards Th 2 cells due to inhibition of Th 1 and Th 9.[47-49]

CHRONIC HEPATITIS C INFECTION AND VITAMIN D

Vitamin D inhibits fibrosis and modifies the natural and acquired immunity by increasing the synthesis of antimicrobial substances and decreases inflammatory cytokines. Many studies have shown that vitamin D deficiency increases the risk of getting influenza, HIV, and respiratory infections.

FIGURE-6-VITAMIN D AND HEPATITIS C



HCV is the main cause of chronic liver disease, it affects the millions of people world wide. Many of them develop cirrhosis and hepatic carcinoma. Significant members of patients will be having lower vitamin D levels and around 25% have severe vitamin D deficiency.

It has been observed that HCV causes suppression of 25 (OH) D levels by means of interrupting lipid pathways as evident by decreasing the production of 7 dehydroxy cholesterol a precursor of Vitamin D .It also causes defective 25 hydroxylation through cytokine induction by oxidative stress .The improved response to treatment with IFN and RBV after Vitamin D supplementation signifies the anti inflammatory action of vitamin D .Pro inflammatory cytokines and chemokines increase the persistence of HCV infection.

Low Th 1/Th 2 ratio determine the sustained viral response rate (SVR) in the treatment of genotype I HCV.[50]As previously told 1-alpha 25 (OH)2 D favours TH 2 production. Vitamin D has anti viral property against HCV infection which is proven by many in vitro studies. Many clinical studies described there exists a relationship between VDR polymorphisms and the treatment of chronic hepatitis C.[51] Gal Tanamy et al study describe that vitamin D inhibits the replication of HCV in liver cells by increasing the VDR expression and IFN B and IFN gene stimulation.

The same study also proves vitamin D supplementation along with anti viral treatment leads to inhibition of HCV.

Vitamin D deficiency is associated with increased necrosis and fibrosis . Poor SVR to interferon therapies is reported in many studies. The relevance of vitamin D in chronic hepatitis C is further proved by the evidence of 50% of the patients with normal vitamin D levels showed improved SVR ,but the others with severe vitamin D deficiency had no SVR .

Bitteto et al gave RS 12979860 C /T polymorphism of IL28B for determining SVR in chronic hepatitis C patients with genotype 1,4,5 which are very difficult to treat.[52] In the study CYP27B1-1260 polymorphism will also decrease the calcitriol in the mononuclear and T lymphocytes cells.[53]

Study conducted by lange and collegues also showed poor SVR in the HCV 1,2,3, genotype infected patients with these polymorphisms.[54]

Vitamin D Deficiency causes insulin resistance which is the important determining factor in the treatment response to IFN and RBV. [55-57]Vitamin D supplementation improves insulin sensitivity and prevents diabetes mellitus type 2 .

VDR is also present in the pancreatic cells .Vitamin D deficiency alters the balance between intra and extra cellular calcium.[58]

TABLE-2-GENETIC VARIATION IN VITAMIN D AND DISEASES

Target	Action
Hepatic	
	1α-hydroxylase (*CYP27B1*) gene:
	rs10877012 A/C SNP associated with responsiveness to therapy in HCV-1
	AA genotype has higher SVR rate and 1α,25(OH)$_2$D level than AC or CC genotype [29]
	VDR gene polymorphism associations:
	bAt [CCA]-haplotype of *BsmI*, *ApaI* and *TaqI* alleles, and CC genotype of the *ApaI* allele predicts rapid fibrosis progression, cirrhosis and increased intrahepatic expression of fibrosis marker gene *MMP-9* [39]
	HBV: eAg positivity and flare [67], higher viral load, more severe fibrosis and necroinflammation [68], chronic infection [69] and occult infection [70]
	BsmI-ApaI-TaqI A-T-C and G-T-T haplotypes associated with HCC in alcohol-related cirrhosis [66]
	BsmI and PBC [71,72]
	FokI and AIH [64,73]
	BsmI-TaqI G-T/G-T diplotype predicts acute rejection post-liver transplantation [74]
Extra-hepatic	
	VDR gene polymorphism associations:
	Immune-mediated diseases: type 1 diabetes [75], leprosy [76], Crohn's disease [77], TB [69,78,100], psoriasis [79], MS [80] and Graves' disease [81]
	Malignancies: melanoma [148] and cancer of the colon [146], ovary [147], breast, prostate and kidney [148]
	DBP gene associations:
	Gc1F, Gc1S and Gc2 isoforms of DBP have differing affinities for vitamin D [133,134] and result in variable responses to vitamin D supplementation [132,133]
	Vitamin D dependent antimicrobial response of monocytes varies with DBP genotype [96]
	Gc2 isoform associated with lower 25(OH)D level [97,98], reduced macrophage function [4] and increased susceptibility to active TB in the presence of severe vitamin D deficiency [99]

TABLE-3-CHILD PUGH CLASSIFICATION

		Points Toward Total Score		
Factor	**Units**	**1**	**2**	**3**
Serum bilirubin	µmol/L	<34	34–51	>51
	mg/dL	<2.0	2.0–3.0	>3.0
Serum albumin	g/L	>35	30–35	<30
	g/dL	>3.5	3.0–3.5	<3.0
Prothrombin time	seconds prolonged	<4	4–6	>6
	INR[a]	<1.7	1.7–2.3	>2.3
Ascites		None	Easily controlled	Poorly controlled
Hepatic encephalopathy		None	Minimal	Advanced

[a]International normalized ratio.

TABLE-4-MELD SCORE

Model for End Stage Liver Disease (MELD) Score

MELD = 3.78 x $\log_e$ serum bilirubin (mg/dL) +
11.20 x $\log_e$ INR +
9.57 x $\log_e$ serum creatinine (mg/dL) +
6.43 (constant for liver disease etiology)

NOTES:

- If the patient has been dialyzed twice within the last 7 days, then the value for serum creatinine used should be 4.0
- Any value less than one is given a value of 1 (i.e. if bilirubin is 0.8, a value of 1.0 is used) to prevent the occurrence of scores below 0 (the natural logarithm of 1 is 0, and any value below 1 would yield a negative result)

VITAMIN D AND NAFLD

NAFLD is a pathological phenomenon indicating a wide range of liver problems from steatosis to non alcoholic steatohepatitis (NASH) and cirrhosis.[59]

NAFLD is one of the important causes of chronic liver disease in developed nations, upto 20% to 30% of the population are affected. Some NAFLD patients gets NASH and cirrhosis, while many others do not go for worsening of the disease ; however, the cause for these differences in worsening of the disease are not known.

NAFLD is commonly associated to at least one metabolic syndrome feature; including liver conditions which are also considered as a part of the syndrome, and though their pathogenesis is not clearly known, IR is an important determining factor in its disease process. Many studies reveal a negative association between vitamin D levels and obesity, glucose intolerance, Insulin resistance, metabolic syndrome and body mass index (BMI).[60,61] And also vitamin D deficiency induces PTH, which has been related to IR and an elevation of the acute-phase reactant.[62-64]

Few researches show that vitamin D supplementation increases secretion of insulin and its use decreases IR in patients with end-stage renal disease. Mostly, VDR polymorphisms have been related to IR and have an effect on secretion of insulin and on the fasting blood glucose levels.[65] The development of hepatic steatosis was observed in VDR knock-out mice in many of the previous studies. Activation of the fibroblastic intestinal growth factor 15 (FGF15) have been shown in many studies by giving vit D supplementation in mice. This intestinal hormone

inhibits CYP7A1 and prevents IR and high-fat diet-induced obesity , an essential enzyme in the pathophysiology of dyslipidemia in the liver.

There is evidence suggesting that vitamin D is associated to the NAFLD development by its use in metabolism of glucose by increasing the change of proinsulin to insulin, whereas vitamin D deficiency has been associated with dysfunction of pancreatic β cell and an increased prevalence of type 2 DM .

Like the case of CHC, vitamin D levels are lesser in patients with NAFLD as compared to healthy controls. And also, in obese patients the accumulation of the vitamin D in adipose tissue has been attributed to vitamin D deficiency[66-68] vitamin D levels has inverse correlation with the worsening of steatosis, necro inflammation and fibrosis and not dependent of age gender, BMI, Homeostatic Model Assesment of IR score and presence of metabolic syndrome.[69,70]

In a recently published clinical article of adults with NAFLD, Targher *et al* showed that the levels of vitamin D will have an effect on hepatic steatosis development and in the histological lesion severity. In fact, their patients with greater inflammation and fibrosis had lower vitamin D levels independent of the presence of other features of the metabolic syndrome. This view was confirmed in pediatric patients later. Even though there exists an association between vitamin D and NAFLD which is not dependent of BMI or IR and other components of metabolic syndrome. Though conclusions are difficult to obtain from these studies, their observations states that vitamin D deficiency plays a important role in the progression and development of fatty liver, in t its anti-inflammatory potential. And also vitamin D in healthy men

reduces the risk for NAFLD and attenuates high fat diet-induced hepatic steatosis in rats by changing lipid metabolism.

Vitamin D deficiency has been associated to a increase in inflammation systemic markers, and this inflammation plays a major role in the pathophysiology and worsening of NAFLD.[71,72] Increase in visceral adipose tissue accelerates the release of pro inflammatory cytokines and fatty acids and stimulates the inflammatory pathways in the liver, prompting pro inflammatory cytokine secretion leading on to damage of the liver.[73] Moreover, in the obese patients the onset of NAFLD is promoted due to excess free fatty acids which causes increased hepatic lipid synthesis followed by; subsequently increasing the oxidative stress on mitochondria and also increase the pro inflammatory cytokines which can surely stimulate the progression of steatosis to NASH and cirrhosis.

In vivo and in vitro studies have precisely stated that steatosis causes reduction in the oxidative activity which is coordinated by cytochrome P450. These process of inflammation is blocked by increasing the 25(OH)D levels.[74] and progression and the development of NAFLD may be halted. vitamin D administration have been shown to decrease markers of inflammation and elevates anti-inflammatory cytokines. It is evident that effect of vitamin D in the liver are exerted not only on the hepatocytes, provided that these cells express very minimal VDR mRNA. In contrast, sinusoidal cells, kupffer cells, hepatic stellate cells and cells of the immune system express VDR mRNA that is functionally active. Therefore, vitamin D deficiency may affect the activity/expression of macrophages, dendritic cells and T

and B lymphocytes by causing oxidative stress and the production of pro inflammatory cytokines leading on to subclinical inflammation.

Further, fibrosis is stimulated by secretion of TGF-β resulting in the increase of secretion of the matrix metalloproteinase 9 inhibitor (TIMP-1). In deed, cell cultures show that vitamin D has an an antifibrinolytic and anti-inflammatory effect on hepatic stellate cells. Finally, animal models show that severity of histological lesions of NAFLD are related to the increased levels of mRNA of TLR2, 4 and 9, proinflammatory cytokines and oxidative stress markers in rats with vitamin D deficiency and a high-fat diet. [75]

A recent study of experimentally NAFLD induced rats showed that exposure to ultraviolet light radiation decreases activity of hepatic stellate cell and TGF-β synthesis and stimulates the production of apolipoprotein E and adiponectin. Altogether, these findings translates into a beneficial effect on NAFLD, and a decrease in IR, steatosis, apoptosis, inflammation and intrahepatic fibrosis was hypothesized. And , given the above-mentioned findings, we can conclude that extrahepatic signaling affects fibrosis and inflammation and that the vitamin D-VDR axis may play a role in the initiation and progression of NAFLD.

Thus, even though the mechanisms of vitamin D's control over hepatic lipid homeostasis and its association with inflammation are not fully known, recent research studies provide a more wholesome understanding of its immune modulatory capacity and of new therapeutic interventions.

CIRRHOSIS

It is usually defined histopathologically .But indicates a variety of clinical manifestations and complications, which can be life-threatening. In the past, cirrhosis was never reversible and it has been thought like that; but, it has become evident that the underlying insult which has caused the cirrhosis is removed, there will be a reversal of fibrosis. This is most evident with the successful treatment of chronic hepatitis C; but fibrosis reversal is also seen in patients with hemochromatosis who have been treated successfully and also in patients with alcoholic liver disease who have stopped alcohol use.

Despite the cause of cirrhosis, the pathology consists ,of the development of fibrosis to the point that there is architectural distortion with the formation of regenerative nodules. It results in a reduction of the hepatocellular mass, and its function, and an alteration in the blood flow. The stimulation of fibrosis occurs with hepatic stellate cells activation, resulting in the production of increased amounts of collagen and other parts of the extracellular matrix.

Clinical manifestations of cirrhosis are due to the result of pathologic changes and increase liver disease severity. Most hepato pathologists gives an assessment of grading and staging when evaluating liver biopsy samples. These grading and staging schemes differ between disease states and have been proposed for most conditions, including chronic viral hepatitis, nonalcoholic fatty liver disease, and primary biliary cirrhosis.

Severe fibrosis usually indicates bridging fibrosis with nodularity designated as stage 3 and cirrhosis designated as stage 4. Patients having cirrhosis have different degrees of compensated liver function, and clinicians need to identify between those who is stable, compensated cirrhosis and those who is decompensated . Patients developing complications of the liver disease and with decompensation should be considered for liver transplantation.

Most of the cirrhosis complications requires specific therapy. Portal hypertension is a significant complicating feature of decompensated cirrhosis and is responsible for the development of ascites and bleeding from esophagogastric varices, two complications that signify decompensated cirrhosis. Loss of hepatocellular function results in jaundice, coagulation disorders, and hypoalbuminemia and contributes to the causes of porto systemic encephalopathy.

The complications of cirrhosis are basically the same regardless of the etiology. Nonetheless, it is useful to classify patients by the cause of their liver disease ; patients can be divided into broad groups with alcoholic cirrhosis, cirrhosis due to chronic viral hepatitis, biliary cirrhosis, and other, less common causes such as cardiac cirrhosis, cryptogenic cirrhosis, and other miscellaneous causes.

ALCOHOLIC CIRRHOSIS

Excessive drinking of alcohol chronically causes different types of chronic liver disease, like alcoholic fatty liver, alcoholic hepatitis, and alcoholic cirrhosis. And also, use of excessive alcohol contributes to liver damage in patients with other liver diseases, like hepatitis C, hemochromatosis, and fatty liver disease caused by obesity.

Chronic alcoholics will be having fibrosis in the absence of additional inflammation and/or necrosis. Fibrosis can be centrilobular, pericellular, or periportal. When fibrosis reaches a certain degree, there is disruption of the normal liver architecture and replacement of liver cells by regenerative nodules. In alcoholic cirrhosis, the nodules are usually <3 mm in diameter; this form of cirrhosis is referred to as micronodular.

With cessation of alcohol use, larger nodules may form, resulting in a mixed micronodular and macronodular cirrhosis.

Alcohol is the most commonly used drug in the United States, and more than two-thirds of adults drink alcohol each year. Thirty percent have had a binge within the past month, and over 7% of adults regularly consume more than two drinks per day.

Unfortunately, more than 14 million adults in the United States meet the diagnostic criteria for alcohol abuse or dependence.

In the United States, chronic liver disease is the tenth most common cause of death in adults, and alcoholic cirrhosis accounts for approximately 40% of deaths due to cirrhosis.

Ethanol is mainly absorbed by the small intestine and, to a lesser degree, through the stomach. Gastric alcohol dehydrogenase (ADH) initiates alcohol metabolism. Three enzyme systems account for metabolism of alcohol in the liver. These include cytosolic ADH, the microsomal ethanol oxidizing system (MEOS), and peroxisomal catalase.

The majority of ethanol oxidation occurs via ADH to form acetaldehyde, which is a highly reactive molecule that may have multiple effects. Ultimately, acetaldehyde is metabolized to acetate by aldehyde dehydrogenase (ALDH). Intake of ethanol increases intracellular accumulation of triglycerides by increasing fatty acid uptake and by reducing fatty acid oxidation and lipoprotein secretion.

Protein synthesis, glycosylation, and secretion are impaired. Oxidative damage to hepatocyte membranes occurs due to the formation of reactive oxygen species; acetaldehyde is a highly reactive molecule that combines with proteins to form protein-acetaldehyde adducts. These adducts may interfere with specific enzyme activities, including microtubular formation and hepatic protein trafficking.

With acetaldehyde-mediated hepatocyte damage, certain reactive oxygen species can result in Kupffer cell activation. As a result, profibrogenic cytokines are produced that initiate and perpetuate stellate cell activation, with the resultant production of excess collagen and extracellular matrix. Connective tissue appears in both periportal and pericentral zones and eventually connects portal triads with central veins forming regenerative nodules.

Hepatocyte loss occurs, and with increased collagen production and deposition, together with continuing hepatocyte destruction, the liver contracts and shrinks in size. This process generally takes from years to decades to occur and requires repeated insults.

The diagnosis of alcoholic liver disease requires an accurate history regarding both amount and duration of alcohol consumption. Patients with alcoholic liver disease can present with nonspecific symptoms such as vague right upper quadrant abdominal pain, fever, nausea and vomiting, diarrhea, anorexia, and malaise.

Alternatively, they may present with more specific complications of chronic liver disease, including ascites, edema, or upper gastrointestinal (GI) hemorrhage. Many cases present incidentally at the time of autopsy or elective surgery. Other clinical manifestations include the development of jaundice or encephalopathy. The abrupt onset of any of these complications may be the first event prompting the patient to seek medical attention. Other patients may be identified in the course of an evaluation of routine laboratory studies that are found to be abnormal. On physical examination, the liver and spleen may be enlarged, with the liver edge being firm and nodular. Other frequent findings include scleral icterus, palmar erythema , spider angiomas , parotid gland enlargement, digital clubbing, muscle wasting, or the development of edema and ascites.

Men may have decreased body hair and gynecomastia as well as testicular atrophy, which may be a consequence of hormonal abnormalities or a direct toxic effect of alcohol on the testes. In women with advanced alcoholic cirrhosis, menstrual irregularities usually occur, and some women may be amenorrheic. These changes are often reversible following cessation of alcohol.

Laboratory tests may be completely normal in patients with early compensated alcoholic cirrhosis. Alternatively, in advanced liver disease, many abnormalities usually are present. Patients may be anemic either from chronic GI blood loss, nutritional deficiencies, or hypersplenism related to portal hypertension, or as a direct suppressive effect of alcohol on the bone marrow. A unique form of haemolytic anemia (with spur cells and acanthocytes) called Zieve's syndrome can occur in patients with severe alcoholic hepatitis. Platelet counts are often reduced early in the disease, reflective of portal hypertension with hypersplenism.

Serum total bilirubin can be normal or elevated with advanced disease. Direct bilirubin is frequently mildly elevated in patients with a normal total bilirubin, but the abnormality typically progresses as the disease worsens. Prothrombin times are often prolonged and usually do not respond to administration of parenteral vitamin K. Serum sodium levels are usually normal unless patients have ascites and then can be depressed, largely due to ingestion of excess free water. Serum alanine and aspartate amino transferases (ALT, AST) are typically elevated, particularly in patients who continue to drink, with AST levels being higher than ALT levels, usually by a 2:1 ratio.

Patients who have any of the above-mentioned clinical features, physical examination findings, or laboratory studies should be considered to have alcoholic liver disease.

The diagnosis, however, requires accurate knowledge that the patient is continuing to use and abuse alcohol. Furthermore, other forms of chronic liver disease (e.g.,chronic viral hepatitis or metabolic or autoimmune liver diseases) must be considered or ruled out, or if present, an estimate of relative causality along with the alcohol use should be determined. Liver biopsy can be helpful to confirm a diagnosis, but generally when patients present with alcoholic hepatitis and are still drinking, liver biopsy is withheld until abstinence has been maintained for at least 6 months to determine residual, non reversible disease.

In patients who have had complications of cirrhosis and who continue to drink, there is a <50% 5-year survival. In contrast, in patients who are able to remain abstinent, the prognosis is significantly improved. In patients with advanced liver disease, the prognosis remains poor; however, in individuals who are able to remain abstinent, liver transplantation is a viable option.

TREATMENT

Abstinence is the cornerstone of therapy for patients with alcoholic liver disease. In addition, patients require good nutrition and long term medical supervision to manage underlying complications that may develop. Complications such as the development of ascites and edema, variceal hemorrhage, or portosystemic encephalopathy all require specific management and treatment.

Glucocorticoids are occasionally used in patients with severe alcoholic hepatitis in the absence of infection.

Survival has been shown to improve in certain studies. Treatment is restricted to patients with a discriminant function (DF) value of >32. The DF is calculated as the serum total bilirubin plus the difference in the patient's prothrombin time compared to control (in seconds) multiplied by 4.6.

In patients for whom this value is >32, there is improved survival at 28 days with the use of glucocorticoids.

Other therapies that have been used include oral pentoxifylline, which decreases the production of tumor necrosis factor α (TNF-α) and other proinflammatory cytokines.

In contrast to glucocorticoids, with which complications can occur, pentoxifylline is relatively easy to administer and has few, if any, side effects. A variety of nutritional therapies have been tried with either parenteral or enteral feedings; however, it is unclear whether any of these modalities have significantly improved survival.

Recent studies have used parenterally administered inhibitors of TNF-α such as infliximab or etanercept. Early results have shown no adverse events; however, there was no clear-cut improvement in survival. Anabolic steroids, propylthiouracil, antioxidants, colchicine, and penicillamine have all been used but do not show clear-cut benefits and are not recommended.

As mentioned above, the cornerstone to treatment is cessation of alcohol use. Recent experience with medications that reduce craving for alcohol, such as acamprosate calcium, has been favorable. Patients may take other necessary medications even in the presence of cirrhosis. Acetaminophen use is often discouraged in patients with liver disease; however, if no more than 2 g of acetaminophen per day are consumed, there generally are no problems.

CIRRHOSIS DUE TO CHRONIC VIRAL HEPATITIS B OR C

Of patients exposed to the hepatitis C virus (HCV), approximately 80% develop chronic hepatitis C, and of those, about 20–30% will develop cirrhosis over 20–30 years.

Many of these patients have had concomitant alcohol use, and the true incidence of cirrhosis due to hepatitis C alone is unknown. Nonetheless, this represents a significant number of patients. It is expected that an even higher percentage will go on to develop cirrhosis over longer periods of time. In the United States, approximately 5 to 6 million people have been exposed to HCV, with about 4 million who are chronically viremic.

Worldwide, about 170 million individuals have hepatitis C, with some areas of the world (e.g., Egypt) having up to 15% of the population infected. HCV is a noncytopathic virus, and liver damage is probably immune-mediated.

Progression of liver disease due to chronic hepatitis C is characterized by portal-based fibrosis with bridging fibrosis and nodularity developing, ultimately culminating in the development of cirrhosis. In cirrhosis due to chronic hepatitis C, the liver is small and shrunken with characteristic features of a mixed, micro- and macronodular cirrhosis seen on liver biopsy. In addition to the increased fibrosis that is seen in cirrhosis due to hepatitis C, an inflammatory infiltrate is found in portal areas with interface hepatitis and occasionally some lobular hepatocellular injury and inflammation.

In patients with HCV genotype 3, steatosis is often present. Similar findings are seen in patients with cirrhosis due to chronic hepatitis B. Of adult patients exposed to hepatitis B, about 5% develop chronic hepatitis B, and about 20% of those patients will go on to develop cirrhosis. Special stains for hepatitis B core (HBc) and hepatitis B surface (HBs) antigen will be positive, and ground-glass hepatocytes signifying hepatitis B surface antigen (HBsAg) may be present.

In the United States, there are about 2 million carriers of hepatitis B, whereas in other parts of the world where hepatitis B virus (HBV) is endemic (i.e., Asia, Southeast Asia, sub- Saharan Africa), up to 15% of the population may be infected, having acquired the infection vertically at the time of birth. Thus, over 300–400 million individuals are thought to have hepatitis B worldwide. Approximately 25% of these individuals may ultimately develop cirrhosis.

Patients with cirrhosis due to either chronic hepatitis C or B can present with the usual symptoms and signs of chronic liver disease. Fatigue, malaise, vague right upper quadrant pain, and laboratory abnormalities are frequent presenting features.

Diagnosis requires a thorough laboratory evaluation, including quantitative HCV RNA testing and analysis for HCV genotype, or hepatitis B ,serologies to include HBsAg, anti-HBs, HBeAg (hepatitis B e antigen), anti-HBe, and quantitative HBV DNA levels.

Management of complications of cirrhosis revolves around specific therapy for treatment of whatever complications occur (e.g., esophageal variceal hemorrhage, development of ascites and edema, or encephalopathy). In patients with chronic hepatitis B, numerous studies have shown beneficial effects of antiviral therapy, which is effective at viral suppression, as evidenced by reducing aminotransferase levels and HBV DNA levels, and improving histology by reducing inflammation and fibrosis. Several clinical trials and case series have demonstrated that patients with decompensated liver disease can become compensated with the use of antiviral therapy directed against hepatitis B.

Currently available therapy includes lamivudine, adefovir, telbivudine, entecavir, and tenofovir. Interferon α can also be used for treating hepatitis B, but it should not be used in cirrhotics. Treatment of patients with cirrhosis due to hepatitis C is a little more difficult because the side effects of pegylated interferon and ribavirin therapy are often difficult to manage.

Dose-limiting cytopenias (platelets, white blood cells, red blood cells) or severe side effects can result in discontinuation of treatment.

Nonetheless, if patients can tolerate treatment, and if it is successful, the benefit is great and disease progression is reduced. Recent studies have shown that if platelets are <100,000, albumin is <3.5 g/dL, and Model for End-Stage Liver Disease (MELD) score is >10, the risk of severe complications of interferon-based antiviral therapy is significant. Recent approval of Direct Acting Antivirals (DAAs) has led to improved efficacy of treatment with regimens that are safe and well tolerated.

CIRRHOSIS FROM AUTOIMMUNE HEPATITIS AND NONALCOHOLIC FATTY LIVER DISEASE

Other causes of posthepatitic cirrhosis include autoimmune hepatitis and cirrhosis due to non alcoholic steatohepatitis. Many patients with autoimmune hepatitis (AIH) present with cirrhosis that is already established. Typically, these patients will not benefit from immunosuppressive therapy with glucocorticoids or azathioprine because the AIH is "burned out." In this situation, liver biopsy does not show a significant inflammatory infiltrate.

Diagnosis in this setting requires positive autoimmune markers such as antinuclear antibody (ANA) or anti-smooth-muscle antibody (ASMA). When patients with AIH present with cirrhosis and active inflammation accompanied by elevated liver enzymes, there can be considerable benefit from the use of immunosuppressive therapy. Patients with nonalcoholic steatohepatitis are increasingly being found to have progressed to cirrhosis. With the epidemic of obesity that continues in Western

countries, more and more patients are identified with nonalcoholic fatty liver disease . Of these, a significant subset has nonalcoholic steatohepatitis and can progress to increased fibrosis and cirrhosis. Over the past several years, it has been increasingly recognized that many patients who were thought to have cryptogenic cirrhosis in fact have nonalcoholic steatohepatitis.

As their cirrhosis progresses, they become catabolic and then lose the telltale signs of steatosis seen on biopsy. Management of complications of cirrhosis due to either AIH or nonalcoholic steatohepatitis is similar to that for other forms of cirrhosis.

COMPLICATIONS OF CIRRHOSIS

The clinical course of patients with advanced cirrhosis is often complicated by a number of important sequelae that can occur regardless of the underlying cause of the liver disease.

These include portal hypertension and its consequences of gastroesophageal variceal hemorrhage, splenomegaly, ascites, hepatic encephalopathy, spontaneous bacterial peritonitis (SBP), hepatorenal syndrome, and hepatocellular carcinoma

Complications of Cirrhosis

Portal hypertension Coagulopathy

Gastroesophageal varices

Factor deficiency

Portal hypertensive gastropathy

Fibrinolysis

Splenomegaly

hypersplenism

Thrombocytopenia

Ascites

Bone disease

Spontaneous bacterial peritonitis

Osteopenia

Hepatorenal syndrome

Osteoporosis

Hepatic encephalopathy

Anemia

Hepatopulmonary syndrome

Hemolysis

Portopulmonary hypertension

Thrombocytopenia

AIM OF THE STUDY

AIMS & OBJECTIVES OF THE STUDY

1) To estimate Vit-25(OH)D level in patients with various types of chronic liver disease.

2) To study the clinical correlation of chronic liver disease patients with hypovitaminosis-D

3) To study the prevalence of vitamin D deficiency in chronic liver disease patients

MATERIALS AND METHODS

CASE DEFINITION

- Definition of Chronic liver disease

Progressive destruction of the liver parenchyma over a period greater than 6 months leading to fibrosis and cirrhosis.

- Hypovitaminosis –D

1.Optimal vitamin D - 30 to 50 ng/mL (i.e. 75-125 nmol/L) .

2.Vitamin D deficiency - lower than 20 ng/mL (i.e. 50 nmol/L) .

3.Vitamin D insufficiency -between 20 and 30 ng/mL (i.e. 50-75nmol/L).

- The relation between hypovitaminosis D and the degree of liver function, degree of fibrosis and infectious complications will be assesed with Child Pugh and MELD scoring criteria.

- **Study design**:-Cross-sectional study

- **Study period**:- 6 months

- **Study area**:- Tertiary health care centre.

- **Study population**:- CLD patients admitted in wards of department of General Medicine .

- **Conflict of Interest** :Nil

SAMPLE SIZE

- The estimated low levels of vitamin-25(OH) D prevalence is 85% in CLD

  with a 10% margin, sample size calculated with a confidence interval of 95% is 55.

  Using the formula 4pq/d2

  p-prevalence q=100-p ,d-marigin of error

INCLUSION CRITERIA

- Patient suffering from chronic liver disease
- Ability to obtain informed consent

EXCLUSION CRITERIA

- Patients suffering from Diabetes mellitus,
- Chronic kidney diseases, Malignancies,
- Ischemic heart diseases and other terminal illnesses.
- Patients on Vitamin D supplements.
- Patients who are all not willing for the study.

METHODOLOGY

- All patients suffering from various types of CLD admitted in medicine wards/ICU with acute worsening of the disease or with any complications or due to progressive worsening of CLD in a tertiary care centre will be included in the study.
- Informed consent will be obtained from the patient /guardian/relatives.
- All patients were subjected to clinical and laboratory evaluation as per proforma
- The patients who are diagnosed as CLD will be categorized based on etiology as alcoholic, nonalcoholic, infective, autoimmune and cryptogenic.
- All the patients will be treated as per institutions CLD protocol throughout hospital stay.
- 6 ml of blood will be collected from each patient at the time of admission.
- Serum vitamin D levels will be measured by ELISA in those patients.
- Complete blood count, RFT, LFT, HBsAg, ANTI-HCV, serum electrolytes, PT-INR, ESR, Ascitic fluid analysis, USG abdomen and Upper GI endoscopy will be done .
- Degree of liver dysfunction will be estimated by Child Pugh Criteria and Model for End stage Liver Disease(MELD) score and outcome will be analysed

DATA COLLECTION AND ANALYSIS

The collected data were analysed with IBM.SPSS statistics software 16.Version.Todescribe about the data descriptive statistics frequency analysis, percentage analysis were used for categorical variables and the mean & S.D were used for continuous variables. To find the association between vitamin D levels and the various study parameters cross tab analysis was done using chi square test ,and the significance difference was calculated using chi square statistic. The association between vitamin D levels with the MELD score and Child Pugh score was calculated using mean, SD and logistic regression test was applied to find out if a statistical difference exists. To find the significance in categorical data Chi-Square test was used similarly if the expected cell frequency is less than 5 in 2×2 tables then the Fisher's Exact was used. In all the above statistical tools the probability value .05 is considered as significant level.

P - Value	** Highly Significant at P ≤ .01
P -Value	# No Significant at P >.05

RESULTS AND OBSERVATIONS

RESULTS AND OBSERVATIONS

DESCRIPTIVE STATISTICS:

NO OF PATIENTS=55

AGE CATEGORY

Mean age of the patients was 45.64 with standard deviation of 6.45.

Minimum age was 20 years and maximum of 63 years.

TABLE-5-AGE CATEGORY

AGE CATEGORY	FREQUENCY	PERCENTAGE
18-35	5	9.1
36-55	46	83.6
>55	4	7.3

In this study there are 46 persons belonging to the age group 36-55.

Total number of patients studied was 55.

5 patients belong to the age group 18-35, 4 patients were above 55 years of age.

83.6% of the patients belong to the age group 36-55.

7.3% belongs to the age group above 55 and 9.1 % belongs to the age group 18-35

FIGURE-7- Bar diagram showing Age Distribution

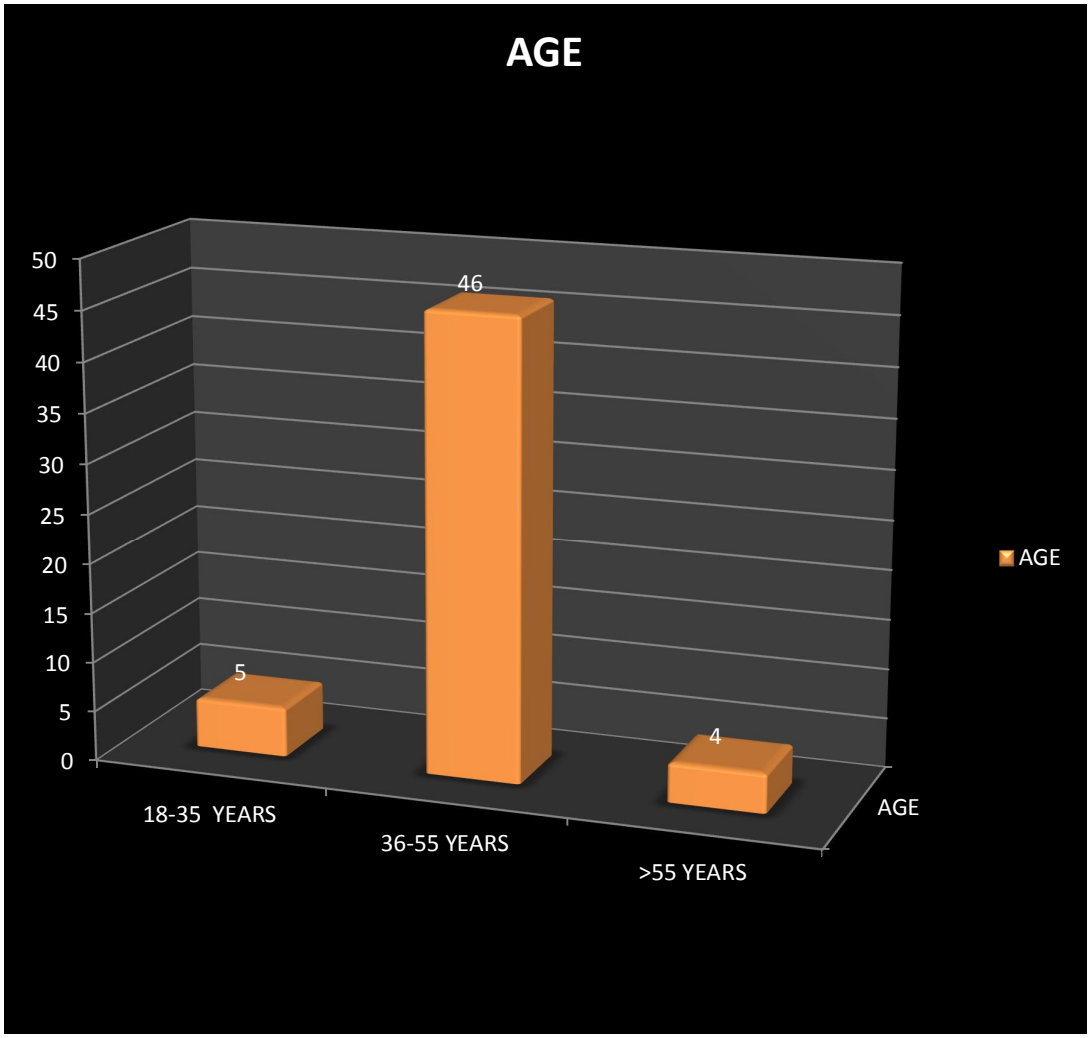
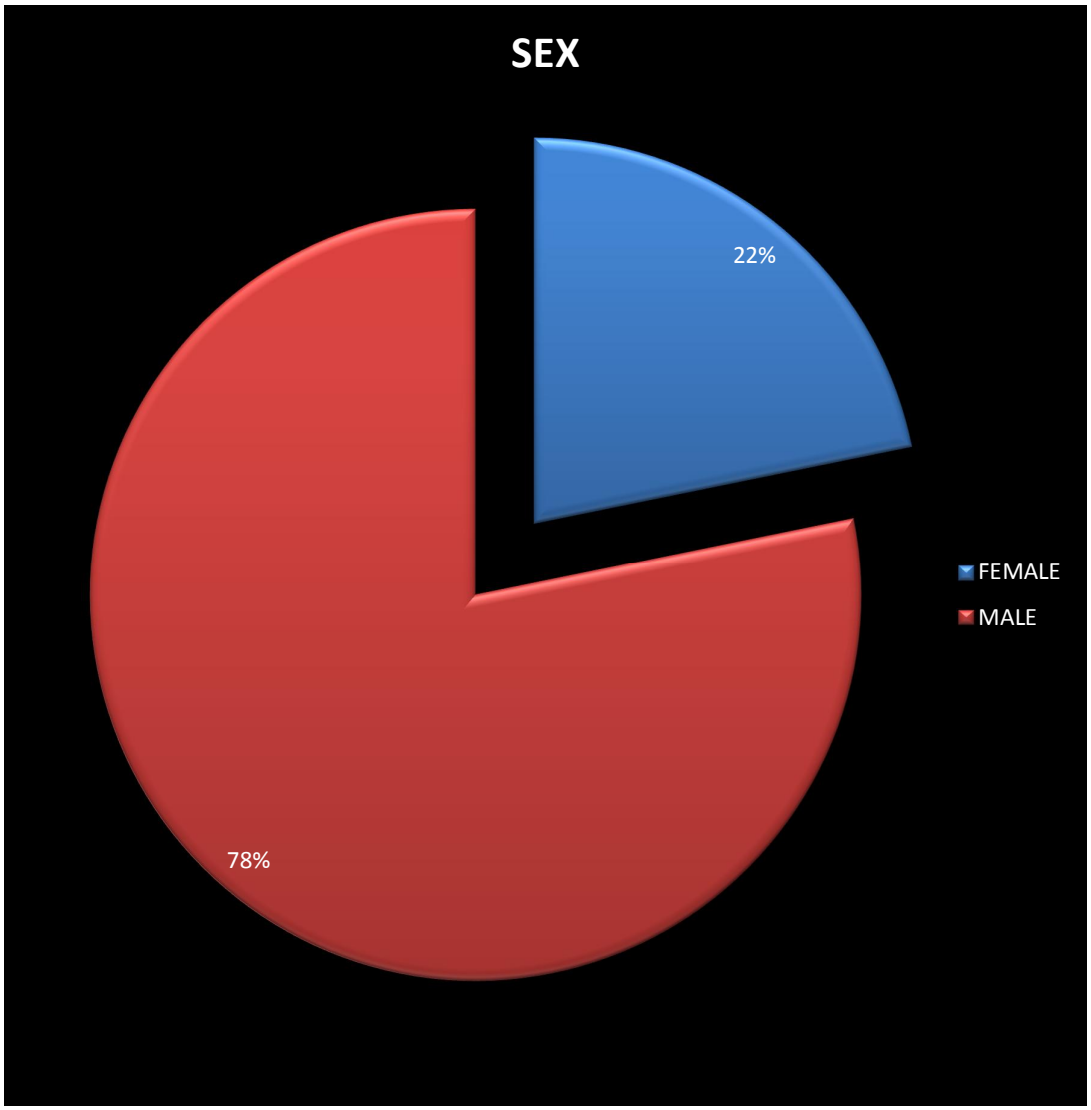


FIGURE -8-Pie chart showing sex distribution



SEX DISTRIBUTION

TABLE-6-SEX DISTRIBUTION

SEX	FREQUENCY	PERCENTAGE
Female	12	21.8
Male	43	78.2

12 patients belongs to female sex group and 43 patients were male patients .

78.2% were male patients and 21.8% were female patients.

BODY MASS INDEX (BMI)

Mean BMI was 23.56 with standard deviation of 3.83.

Minimum BMI was 16.5 and maximum of 32.

TABLE-7-BMI CATEGORY

BMI	FREQUENCY	PERCENTAGE
UNDERWEIGHT	6	10.9
NORMAL	32	58.2
OVERWEIGHT	14	25.5
OBESE	3	5.5

32 patients i.e 58.2% belongs to normal BMI category.

14% belongs to overweight category i.e. around 25.5%.

Whereas 10.9% i.e. 6 patients were underweight and 3 patients were obese with 5.5%.

FIGURE-9-Bar diagram showing BMI category

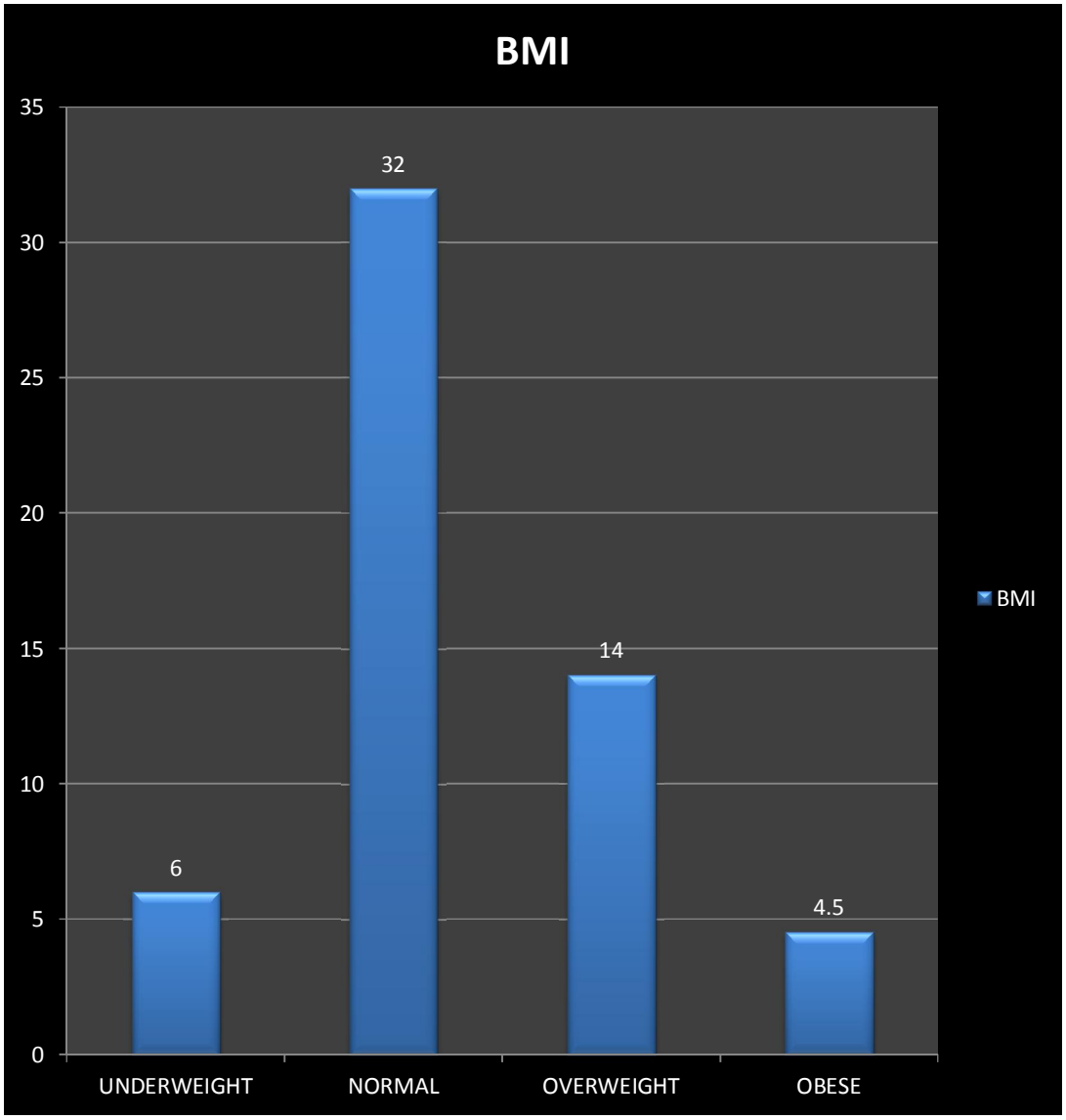
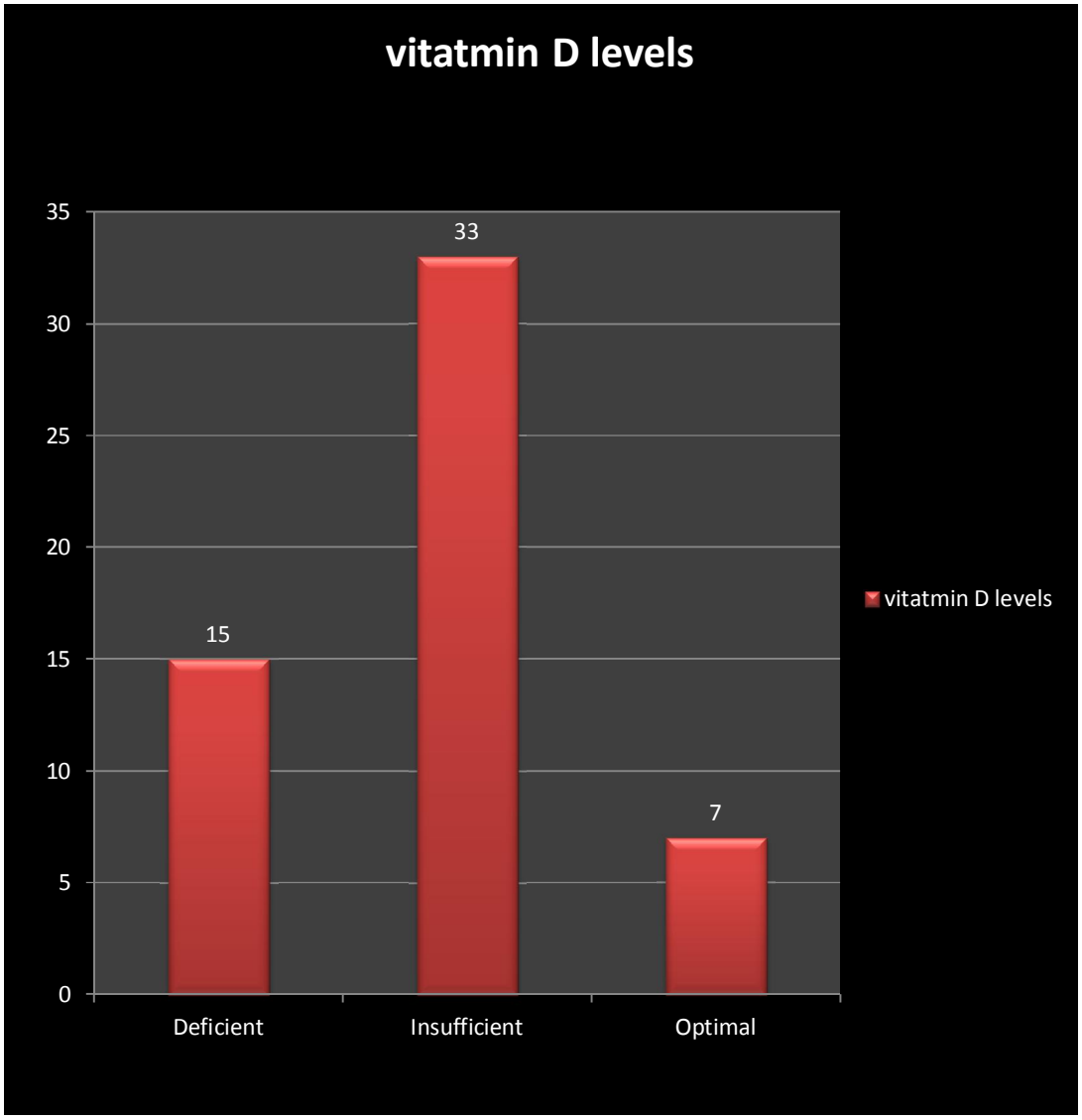


FIGURE-10-Bar diagram showing Vitamin D level in study patients



VITAMIN D LEVEL

Mean vitamin D levels was 23.69 with standard deviation of 6.28.

Minimum level was 10 and maximum was 36.

TABLE-8-VITAMIN D CATEGORY

VITAMIN D CATEGORY	FREQUENCY	PERCENTAGE
DEFICIENT	15	27.3
INSUFFICIENT	33	60
OPTIMAL	7	12.7

Out of 55 patients 15 were having vitamin D levels coming under deficient category.

Insufficient vitamin D Levels was observed in 33 patients and only 7 were having optimal vitamin D levels.

27.3% were deficient, 60%belongs to insufficient category and 12.7% belongs to optimal category.

TOTAL SERUM BILIRUBIN:

Mean serum bilirubin levels was 5.83 with std deviation of 3.01.

Minimum level was 2 and maximum was 12.

SERUM ALBUMIN:

Mean serum albumin levels were 3.215 with standard deviation of 0.56.

Minimum of 2 and maximum of 4.4

SERUM CREATININE:

Mean serum creatinine levels were 1.68 with standard deviation of 1.69.

Minimum of 0.4 and maximum of 7.7

INR:

Mean INR levels were 1.79 with standard deviation of 1.24.

Minimum of 0.8 and maximum of 6.5

PT PROTHROMIN:

Mean PT prothrombin levels were 3.51 with standard deviation of 1.73.

Minimum of 1.5 and maximum of 7.

ASCITES:

TABLE-9-CONTROL OF ASCITES

ASCITES	FREQUENCY	PERCENTAGE
EASILY CONTROLLED	42	76.4
NO	13	23.6

Out of 55 patients 42 were having easily controllable ascites whereas 13 patients have poorly controlled ascities .

76.4% belongs to the easily controlled group .

23.6% belongs to the poorly controllable group.

FIGURE-11-Pie chart showing ascites in view of treatment

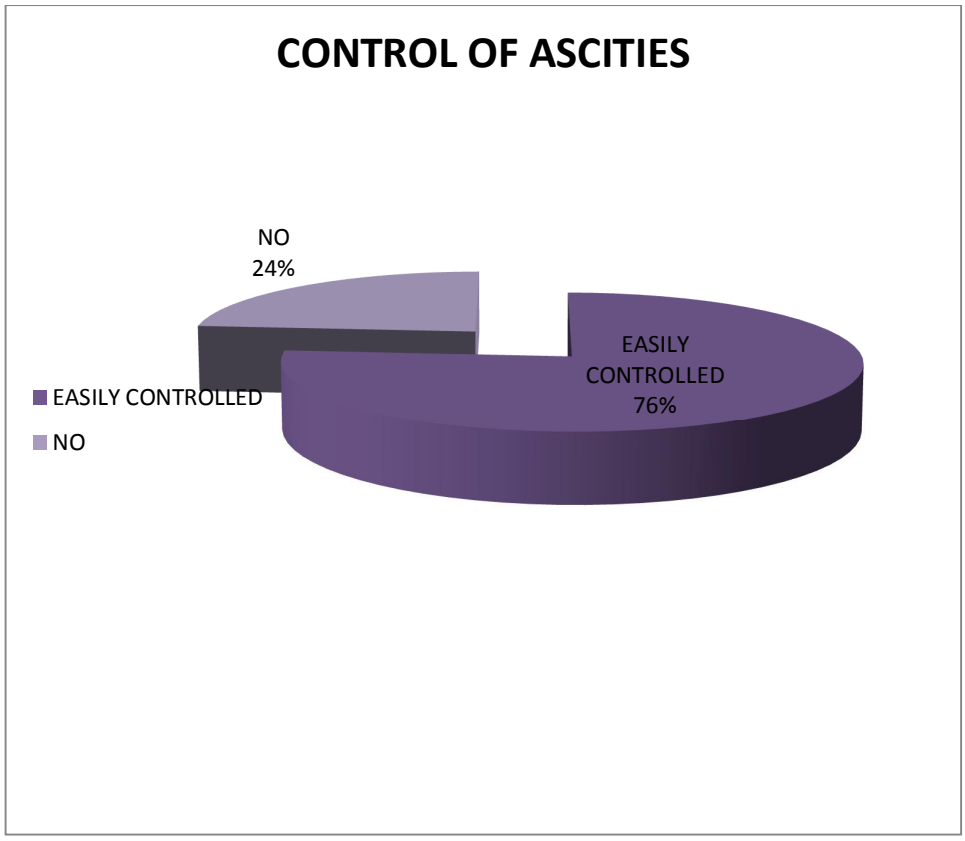
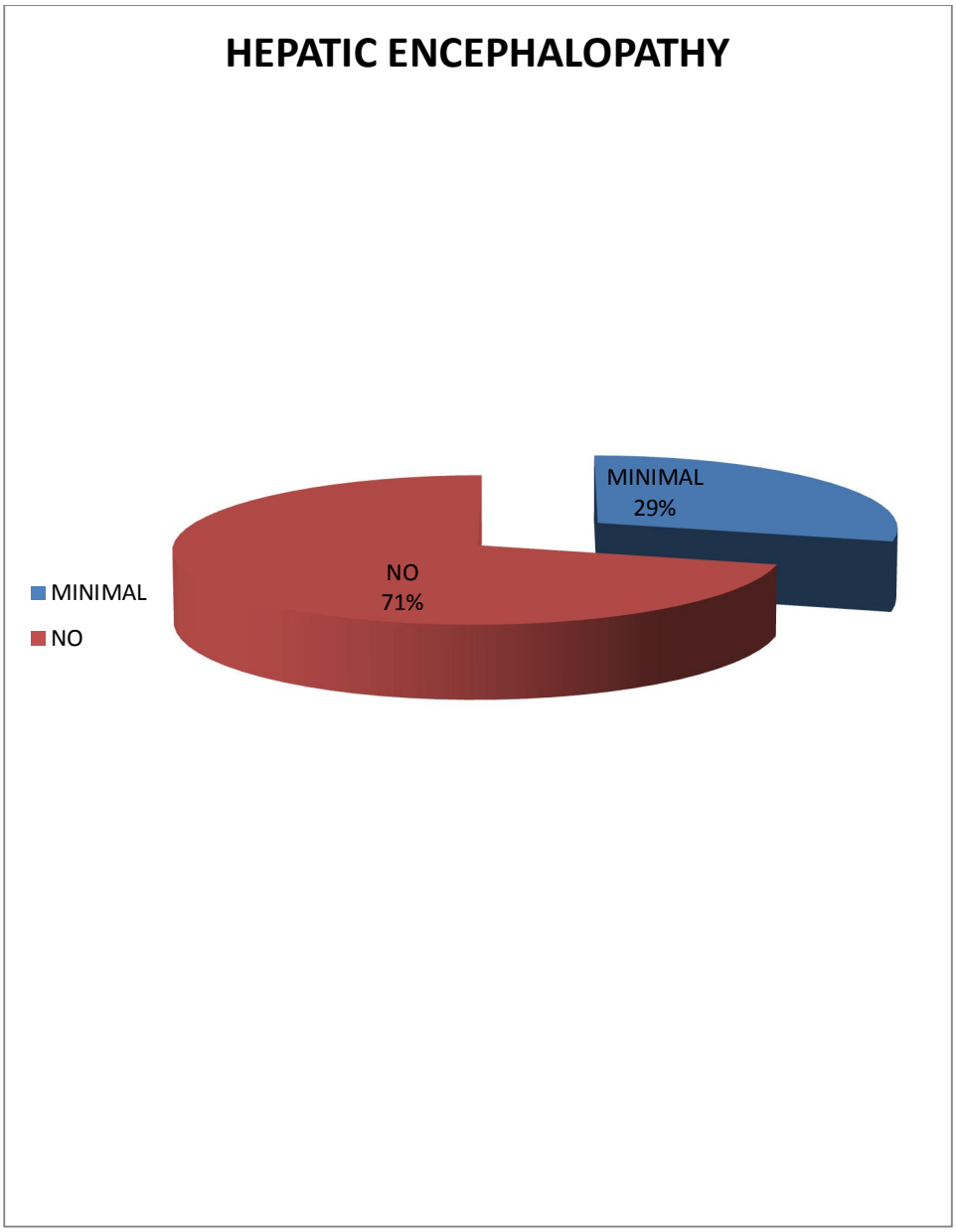


FIGURE-12-pie chart showing heaptic encephalopathy distribution



HEPATIC ENCEPHALOPATHY

TABLE-10-HE DISTRIBUTION IN STUDY PATIENTS

HE	FREQUENCY	PERCENTAGE
MINIMAL	16	29.1
NO	39	70.9

Among the 55 patients 16 were having minimal HE i.e.29.1%

39 were not having hepatic encephalopathy i.e.70.9%

ALCOHOLIC

TABLE-11-ALCOHOLIC ETIOLOGY

ALCOHOLIC	FREQUENCY	PERCENTAGE
YES	43	78.2
NO	12	21.8

In this study 43 patients are alcoholic

12 patients are not an alcoholic

78.2% belongs to the alcohol drinking group.

21.8% belongs to the non alcohol drinking group.

Figure-13-Pie chart showing alcohol distribution in study patients

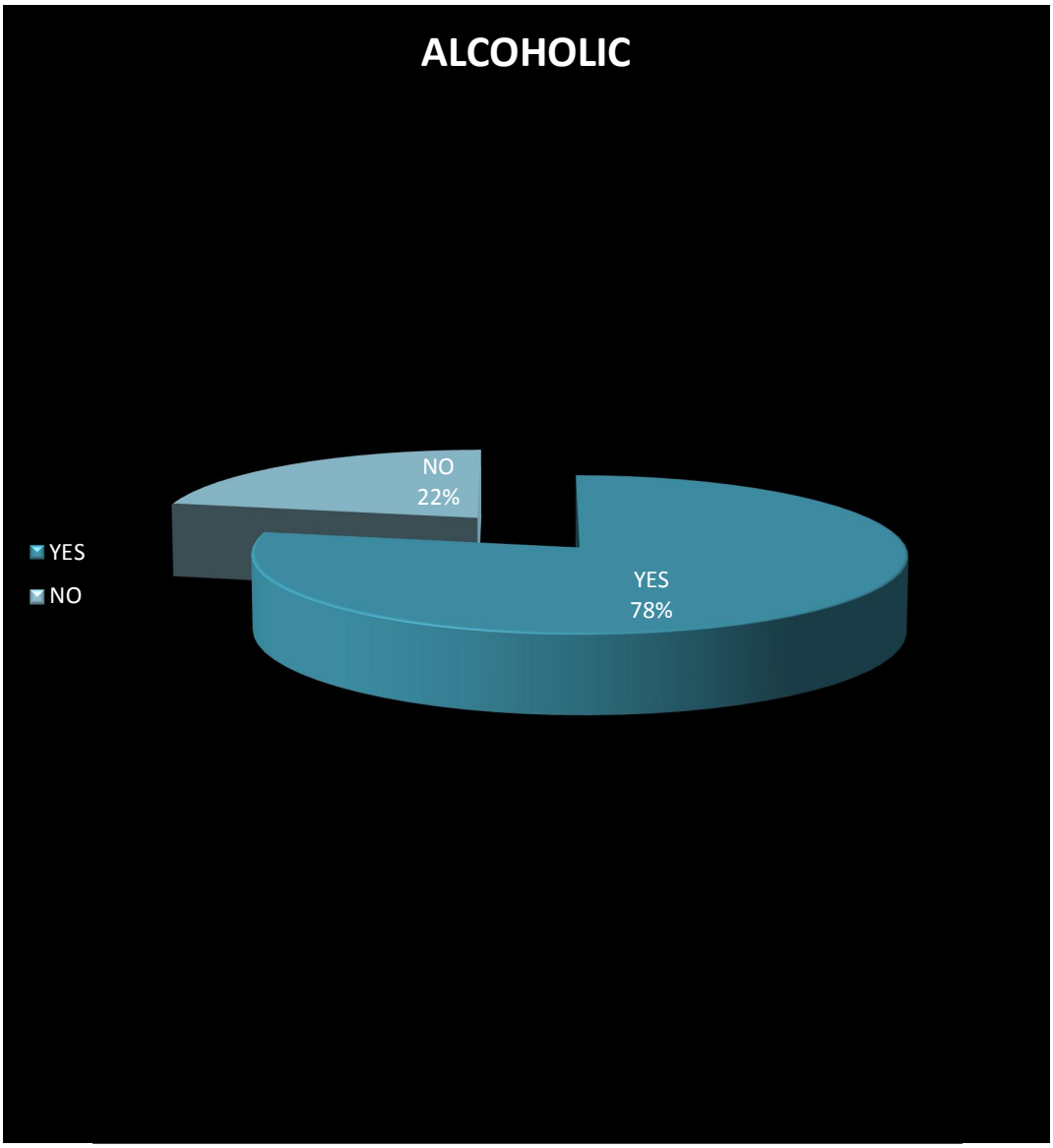
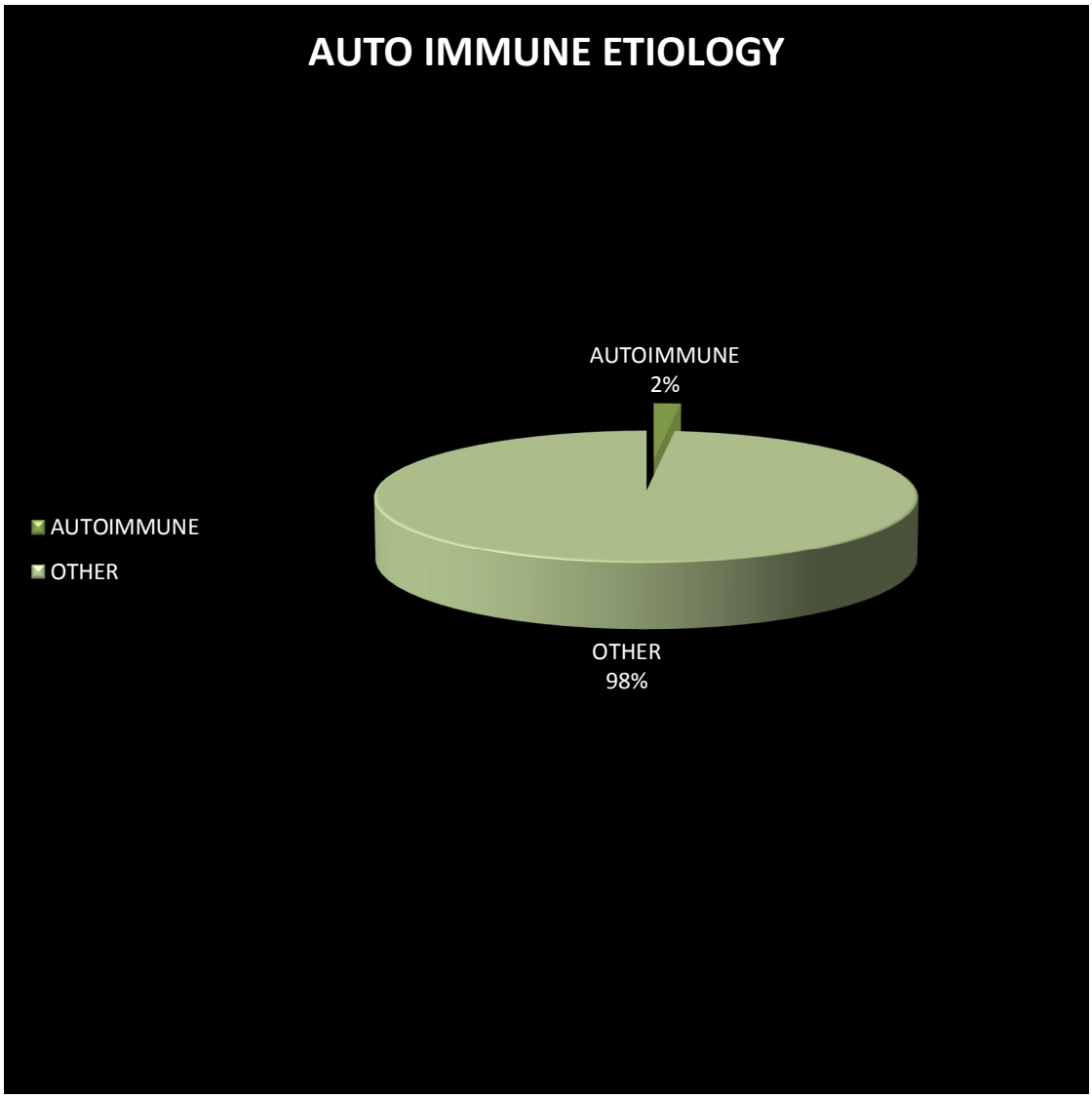


Figure -14-Pie chart showing auto immune Etiology



AUTO IMMUNE ETIOLOGY

Only one i.e. 1.8% patient had an auto immune etiology

whereas 54% i.e.98.2% had other etiologies for chronic liver diseases.

VIRAL ETIOLOGY

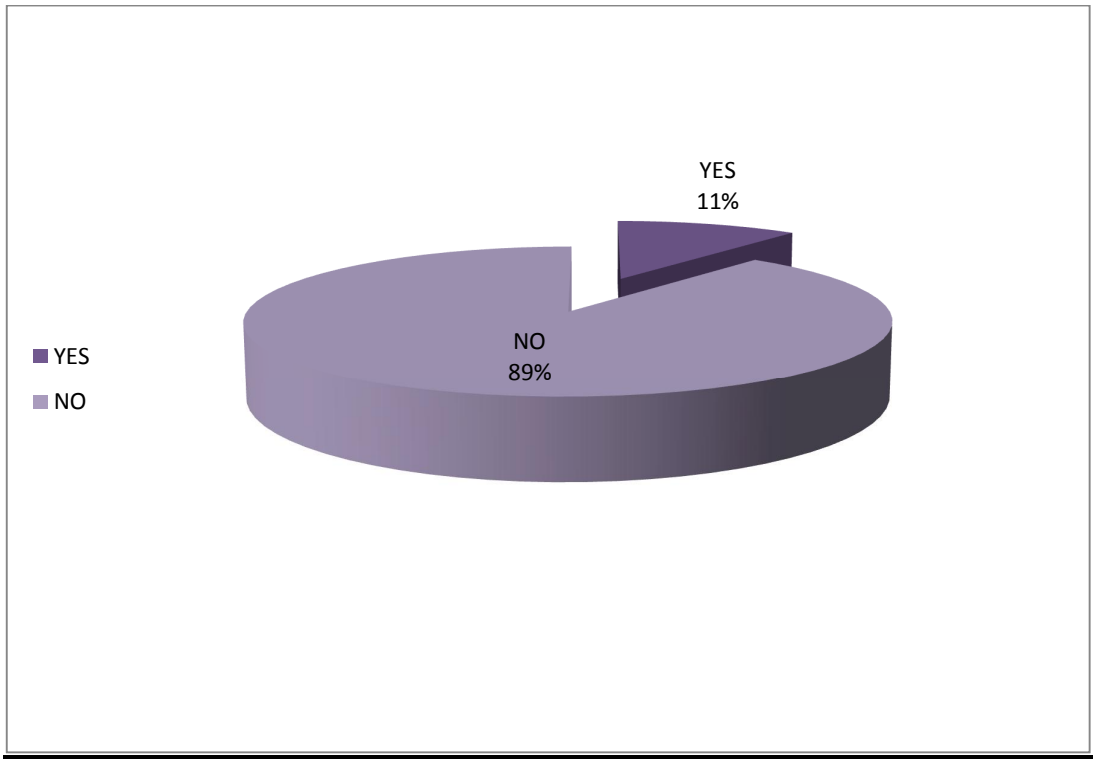
TABLE-12-VIRAL ETIOLOGY AMONG STUDY PATIENTS

VIRAL MARKER	FREQUENCY	PERCENTAGE
NO	49	89.1
YES	6	10.9

Among 55 patients 6 patients i.e. 10.9% have viral aetiology remaining 49 belongs to

Other etiologies.

FIGURE -15-Pie chart showing Viral Etiology

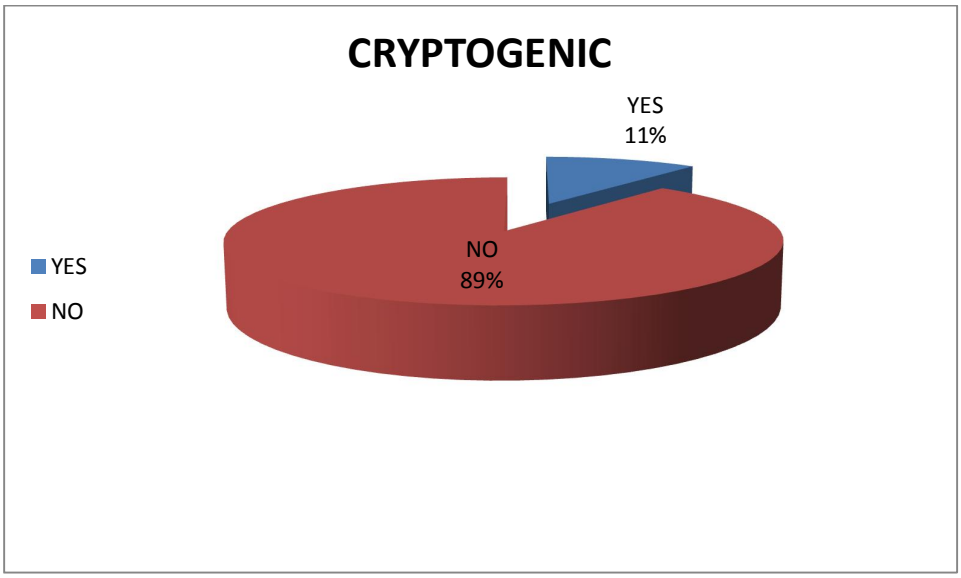


CRYPTOGENIC

TABLE-13-CRYPTOGENIC ETIOLOGY

CRYPTOGENIC	FREQUENCY	PERCENTAGE
YES	6	10.9
NO	49	89.1

FIGURE-16-Pie chart showing cryptogenic etiology



Among 55 patients 6 patients i.e 10.9% have cryptogenic etiology remaining 49 belongs to other etiologies.

RBS

Mean value was 98.25 with std deviation 5.98.

Minimum level was 84 and maximum of 110.

CHILD PUGH SCORE:

Mean score was 9.24 with standard deviation 2.34. Minimum of 6 and maximum of 13

TABLE-14-CHILD PUGH SCORE DISTRIBUTION

CHILD PUGH SCORE	FREQUENCY	PERCENTAGE
A	13	23.6
B	23	41.8
C	19	34.5

Among the study 23 patients belongs to Child Pugh class B and 19 patients belongs to Child Pugh class C.

13 patients belongs to Child Pugh class a i.e.23.6%.

41.8% were Child Pugh class B and 34.5% were child Pugh class C.

MELD SCORE

Mean score was 20.96 with standard deviation 11.85. Minimum of 22.9 and maximum of 71.3

TABLE-15-MELD SCORE DISTRIBUTION

MELD SCORE	FREQUENCY	PERCENTAGE
6%	38	69.1
19.6%	6	10.9
52.6%	5	9.1
71.3%	6	10.9

In this study 38 patients belongs to MELD score of 6% constituting around 69.1%.Whereas 6 patients belong to 19.6% and 71.3% of MELD scoring that constitutes about 10.9% .Whereas 5 patients belongs to 52.6% MELD scoring that constitutes around 9.1%.

TABLE-16-DISTRIBUTION OF VITAMIN D LEVEL

VITAMIN D LEVEL	OPTIMAL	DEFICIENT	INSUFFICIENT	P value
No. of patients (n=55)	7 (12.7%)	15 (27.3%)	33 (60%)	
Sex				
Male (n=43)	5 (11.6%)	12 (27.9%)	26 (60.5%)	P=0.89
Female (n=12)	2 (16.7%)	3 (25%)	7 (58.3%)	
Mean age	41.29 ± 12.64	47.07 ± 4.75	45.91 ± 5.00	
Age (years)				
18-35 (n=5)	3 (60%)	0	2 (40%)	**P=0.02**
36-55 (n=46)	3 (6.5%)	14 (30.4%)	29 (63%)	
>55 (n=4)	1 (25%)	1 (25%)	2 (50%)	
Total	7 (12.7%)	15 (27.3%)	33 (60%)	
Etiology of CLD				
Alcoholic (n=43)	4 (9.3%)	12 (27.9%)	27 (62.8%)	P= 0.41
Ascites (n=42)	0	15 (35.7%)	27 (64.3%)	**P = 0.001**
Viral marker (n=6)	2 (33.3%)	1 (16.7%)	3 (50%)	P = 0.35
Auto-immune (n=1)	1 (100%)	0	0	P = 0.12
Cryptogenic (n=6)	1 (16.7%)	2 (33.3%)	3 (50%)	P= 0.70
HE (n=16)	0	14 (87.5%)	2 (12.5%)	**P = 0.001**

BMI				
UNDERWEIGHT	16.7%(1)	33.3%(2)	50%(3)	**P=0.176**
NORMAL	31.3%(10)	56.3%(18)	12.5%(4)	
OVERWEIGHT	14.3%(2)	50%(7)	35.7%(5)	
OBESE	66.7%(2)	33.3%(1)	0%(0)	

The BMI and vitamin D level were inversely related but no significant association was made out. In alcoholics 27 patients were having insuffcient levels.12 were having deficient levels and 4 were having normal levels. In the viral etiology group 3 were having insufficient levels ,1 deficient,2 were normal. In the autoimmune group only one with normal level was found. In the cryptogenic group 3 were insufficient,2 were deficient,1 were normal.

Similarly there was no significant association between vitamin D levels and age, sex and etiology groups.

There was significant association between MELD and CHILD PUGH score.

TABLE-17-COMPARISON OF MEAN VALUE OF VITAMIN D LEVEL IN RELATION TO MELD SCORE

Parameter	MELD score					P value	Significance
	<9 (n=1)	**10 - 19 (n=37)**	**20 - 29 (n=6)**	**30 - 39 (n=5)**	**>40 (n=6)**		
vitamin D mean + SD	30	26.41 ± 5.22	18.00 ± 4.14	16.00 ± 3.46	18.00 ± 2.00	**<0.001**	**significant**

On Linear regression analysis of vitamin D level we found significant negative correlation with MELD score **(r = – 0.816, P<0.0001).**

There was a significant association between vitamin D levels and severity of chronic liver disease assessed by MELD scoring. There are about 6 patients who have MELD score greater than 40 with vitamin D levels around 18+_2 and 5 patients have MELD score of 30-39 with vitamin D levels 16+_3.46.

6 patients were having MELD of 10-19 with Vitamin D Levels of 18+_4.14 and 37 patients Were having MELD of 10-19 with vitamin D level of 26.4+_5.22 and only 1 patient is having MELD <9 with vitamin D level of 30.

TABLE-18-COMPARISON OF MEAN VALUE OF VITAMIN D LEVEL IN RELATION TO CHILD-PUGH SCORE

Parameter	**Child Pugh score**			**P value**	**significance**
	Class A (5-6) (n=13)	Class B (7-9) (n=23)	Class C(10-15) (n=19)		
vitamin D mean + SD	32.23 ± 2.24	23.65 ± 2.63	17.81 ± 4.29	**<0.001**	**significant**

On Linear regression analysis of vitamin D level we found significant negative correlation with Child-Pugh score (r= –0.875, P<0.0001) .

The average of vitamin D level in association with child pugh scoring was significant with P <0.001 +_2.24 belonging to class A and 23 have vitamin D values at 23.65+_2.63 belonging to class B and 19 patients have vitamin D levels of 17.8%+_4.29 belongs to Class C.

DISCUSSION

DISCUSSION

In our study 55 patients were studied ,mean age of the patient was 45.64 with standard deviation of 6.45 out of which 12 were female (21.8%) and 43 were males (78.2%).Most of them belongs to age group 36-55 years.

In Bal kishan gupta et al study 91% were males and 9% were only females. Putz bankuti c et al study showed 32% females and 68% males similar to our study. Prevalence of female chronic liver disease is towards the increasing trend because of the increasing alcohol drinking habit among the females .

In alcoholics 27 patients were having insuffcient levels.12 were having deficient levels and 4 were having normal levels. In the viral etiology group 3 were having insufficient levels ,1 deficient,2 were normal. In the autoimmune group only one with normal level was found. In the cryptogenic group 3 were insufficient,2 were deficient,1 were normal.

In our study 33(60%) have vitamin D insufficiency out of which 26 were males and 7 were females. And 15(27.3%) have vitamin D deficiency and 12 were males and 3 were females. 7(12.7%) have normal vitamin D levels 5 in males and 2 in females.

Totally 87.3% have subnormal vitamin D levels in our study where as Bal kishan gupta et al showed 85% having subnormal vitamin D levels similar to our study.

On comparison of mean value of Vitamin D level in relation to MELD score by linear regression analysis significant negative correlation with child pugh score ($r=-0.875, P<0.0001$)

So low levels of Vitamin D were associated with increased with severity of the liver disease.

Bal kishan gupta et al showed similar results with child pugh score ($r=-0.7382, p<0.0001$) and MELD score ($r=-0.6673, P<0.0001$).

Putz bankuti et al showed similar results. So the vitamin D deficiency in liver disease is probably because of decreased hydroxylation of vitamin D and decreased vitamin D binding protein production .It is also due to decreased intestinal absorption of vitamin D due to intestinal edema secondary to portal hypertension and cholestasis induced bile salt disruption, jaundice related decreased food intake and poor sun exposure.

Immunomodulatory effect of innate and adaptive immunity also plays a role in causing severity of fibrosis by the decreased expression of VDR and DBP

So our study also proves that vitamin d is associated with prognosis of chronic liver disease implicating it as a biomarker of severity and a efficient therapeutic goal in chronic liver disease.

CONCLUSION

CONCLUSION

We observe a strong association between the lower vitamin D levels and chronic liver disease severity progression.

This association is independent of age ,sex, BMI and etiology.

There is a strong association between lower vitamin D levels and child pugh score.

There is a strong association between lower vitamin D levels and MELD score.

Despite the cross sectional design and limited sample size there is exists a strong contribution of vitamin D deficiency in the worsening of liver diseases.

Further interventional studies by pre and post vitamin D supplementation of chronic liver disease patients will help in treating these chronic liver disease patients in future.

ANNEXURES

BIBLIOGRAPHY

REFERENCES

1. Arteh J, Narra S, Nair S. Prevalence of vitamin D deficiencyin chronic liver disease. *Dig Dis Sci* 2010; 55: 2624-2628 [PMID:19960254 DOI: 10.1007/s10620-009-1069-9]
2. Fisher L, Fisher A. Vitamin D and parathyroid hormone in outpatients with noncholestatic chronic liver disease. *Clin Gastroenterol Hepatol* 2007; 5: 513-520 [PMID: 17222588]
3. Holick MF. Vitamin D deficiency. *N Engl J Med* 2007; 357: 266-281 [PMID: 17634462]
4. DeLuca HF. Overview of general physiologic features and functions of vitamin D. *Am J Clin Nutr* 2004; 80: 1689S-1696S [PMID: 15585789]
5. Yamamoto N, Homma S. Vitamin D3 binding protein (group-specific component) is a precursor for the macrophage- activating signal factor from lysophosphatidylcholine- treated lymphocytes. *Proc Natl Acad Sci USA* 1991; 88:8539-8543 [PMID: 1924312]
6. Metcalf JP, Thompson AB, Gossman GL, Nelson KJ, Koyama S, Rennard SI, Robbins RA. Gcglobulin functions as a cochemotaxin in the lower respiratory tract. A potential mechanism for lung neutrophil recruitment in cigarette smokers.
7. Akeno N, Saikatsu S, Kawane T, Horiuchi N. Mouse vitamin D-24-hydroxylase: molecular cloning, tissue distribution, and transcriptional

regulation by 1alpha,25-dihydroxyvitamin D3. *Endocrinology* 1997; 138: 2233-2240 [PMID: 9165006] *ev Respir Dis* 1991; 143: 844-849 [PMID: 2008995]

8. Messa P, Alfieri C, Rastaldi MP. Recent insights into vitamin D and its receptor. *J Nephrol* 2011; 24 Suppl 18: S30-S37 [PMID: 21623580]
9. Ramagopalan SV, Heger A, Berlanga AJ, Maugeri NJ, Lincoln MR, Burrell A, Handunnetthi L, Handel AE, Disanto G, Orton SM, Watson CT, Morahan JM, Giovannoni G, Ponting CP, Ebers GC, Knight JC. A ChIP-seq defined genome-wide map of vitamin D receptor binding: associations with disease and evolution. *Genome Res* 2010; 20: 1352-1360 [PMID: 20736230 DOI: 10.1101/gr.107920.110].
10. Bookout AL, Jeong Y, Downes M, Yu RT, Evans RM, Mangelsdorf DJ. Anatomical profiling of nuclear receptor expression reveals a hierarchical transcriptional network. *Cell* 2006; 126: 789-799 [PMID: 16923397]
11. Van Belle TL, Gysemans C, Mathieu C. Vitamin D in autoimmune, infectious and allergic diseases: a vital player? *Best Pract Res Clin Endocrinol Metab* 2011; 25: 617-632 [PMID: 21872803]
12. Mora JR, Iwata M, von Andrian UH. Vitamin effects on the immune system: vitamins A and D take centre stage. *Nat Rev Immunol* 2008; 8: 685-698 [PMID: 19172691]
13. Holick MF, Garabedian M. Vitamin D: photobiology, metabolism, mechanism of action, and clinical applications. In: Favus MJ, editor. Primer on the

metabolic bone diseases and disorders of mineral metabolism. Washington, DC: American Society for Bone and Mineral Research, 2006: 129-137

14. Bland R, Walker EA, Hughes SV, Stewart PM, Hewison M. Constitutive expression of 25-hydroxyvitamin D3-1alphahydroxylase in a transformed human proximal tubule cell line: evidence for direct regulation of vitamin D metabolism by calcium. *Endocrinology* 1999; 140: 2027-2034 [PMID: 10218951]
15. Bai XY, Miao D, Goltzman D, Karaplis AC. The autosomal dominant hypophosphatemic rickets R176Q mutation in fibroblast growth factor 23 resists proteolytic cleavage and enhances in vivo biological potency. *J Biol Chem* 2003; 278: 9843-9849 [PMID: 12519781]
16. Schiødt FV. Gc-globulin in liver disease. Dan Med Bull 2008;55:131–146
17. Abu-Mouch S, Fireman Z, Jarchovsky J, Zeina AR, Assy N. Vitamin D supplementation improves sustained virologic response in chronic hepatitis C (genotype 1)-naïve patients. World J Gastroenterol 2011;17:5184–5190.
18. Nimer A, Mouch A. Vitamin D improves viral response in hepatitis C genotype 2–3 naïve patients. World J Gastroenterol 2012;18:800–805.
19. Bitetto D, Fabris C, Fornasiere E, Pipan C, Fumolo E, Cussigh A, et al. Vitamin D supplementation improves response to antiviral treatment for recurrent hepatitis C. Transpl Int 2011;24:43–50.

20. Boonstra A, Barrat FJ, Crain C, Heath VL, Savelkoul HF, O'Garra A. 1a,25-dihydroxyvitamin D3 has a direct effect on naive CD4(+) T cells to enhance the development of Th2 cells. J Immunol 2001;167:4974–4980.

21. Penna G, Adorini L. 1a,25-dihydroxyvitamin D3 inhibits differentiation, maturation, activation, and survival of dendritic cells leading to impaired alloreactive T cell activation. J Immunol 2000;164: 2405–2411.

22. D'Ambrosio D, Cippitelli M, Cocciolo MG, Mazzeo D, Di Lucia P, Lang R, et al. Inhibition of IL-12 production by 1,25-dihydroxyvitamin D3. Involvement of NF-kappaB downregulation in transcriptional repression of the p40 gene. J Clin Invest 1998;101:252–262.

23. Pittas AG, Lau J, Hu FB, Dawson-Hughes B. The role of vitamin D and calcium in type 2 diabetes. A systematic review and meta-analysis. J Clin Endocrinol Metab 2007;92:2017–2029.

24. Trépo E, Ouziel R, Pradat P, Momozawa Y, Quertinmont E, Gervy C, Gustot T, Degré D, Vercruysse V, Deltenre P, Verset L, Gulbis B, Franchimont D, Devière J, Lemmers A, Moreno C. Marked 25-hydroxyvitamin D deficiency is associated with poor prognosis in patients with alcoholic liver disease. *J Hepatol* 2013; 59: 344-350 [PMID: 23557869 DOI: 10.1016/j.jhep.2013.03.024]

25. Putz-Bankuti C, Pilz S, Stojakovic T, Scharnagl H, Pieber TR, Trauner M, Obermayer-Pietsch B, Stauber RE. Association of 25-hydroxyvitamin D levels with liver dysfunction and mortality in chronic liver disease. *Liver Int* 2012; 32: 845-851 [PMID: 22222013 DOI: 10.1111/j.1478-3231.2011.02735.x]

26. White P, Cooke N. The multifunctional properties and characteristics of vitamin D-binding protein. *Trends Endocrinol Metab* 2000; 11: 320-327 [PMID: 10996527]

27. Liu PT, Stenger S, Li H, Wenzel L, Tan BH, Krutzik SR, Ochoa MT, Schauber J, Wu K, Meinken C, Kamen DL, Wagner M, Bals R, Steinmeyer A, Zügel U, Gallo RL, Eisenberg D, Hewison M, Hollis BW, Adams JS, Bloom BR, Modlin RL. Toll-like receptor triggering of a vitamin D-mediated human antimicrobial response. *Science* 2006; 311: 1770-1773 [PMID: 16497887]

28. Sadeghi K, Wessner B, Laggner U, Ploder M, Tamandl D, Friedl J, Zügel U, Steinmeyer A, Pollak A, Roth E, Boltz- Nitulescu G, Spittler A. Vitamin D3 down-regulates monocyte TLR expression and triggers hyporesponsiveness to pathogen-associated molecular patterns. *Eur J Immunol* 2006; 36: 361-370 [PMID: 16402404]

29. Shea MK, Benjamin EJ, Dupuis J, Massaro JM, Jacques PF, D' Agostino RB, Ordovas JM, O'Donnell CJ, Dawson-Hughes B, Vasan RS, Booth SL. Genetic and non-genetic correlates of vitamins K and D. *Eur J Clin Nutr* 2009; 63: 458-464 [PMID: 18030310]

30. Hunter D, De Lange M, Snieder H, MacGregor AJ, Swaminathan R, Thakker RV, Spector TD. Genetic contribution to bone metabolism, calcium excretion, and vitamin D and parathyroid hormone regulation. *J Bone Miner Res* 2001; 16: 371-378 [PMID: 11204437]

31. Wang TJ, Zhang F, Richards JB, Kestenbaum B, van Meurs JB, Berry D, Kiel DP, Streeten EA, Ohlsson C, Koller DL, Peltonen L, Cooper JD, O'Reilly PF,

Houston DK, Glazer NL, Vandenput L, Peacock M, Shi J, Rivadeneira F, McCarthy MI, Anneli P, de Boer IH, Mangino M, Kato B, Smyth DJ, Booth SL, Jacques PF, Burke GL, Goodarzi M, Cheung CL, Wolf M, Rice K, Goltzman D, Hidiroglou N, Ladouceur M, Wareham NJ, Hocking LJ, Hart D, Arden NK, Cooper C, Malik S, Fraser WD, Hartikainen AL, Zhai G, Macdonald HM, Forouhi NG, Loos RJ, Reid DM, Hakim A, Dennison E, Liu Y, Power C, Stevens HE, Jaana L, Vasan RS, Soranzo N, Bojunga J, Psaty BM, Lorentzon M, Foroud T, Harris TB, Hofman A, Jansson JO, Cauley JA, Uitterlinden AG, Gibson Q, Järvelin MR, Karasik D, Siscovick DS, Econs MJ, Kritchevsky SB, Florez JC, Todd JA, Dupuis J, Hyppönen E, Spector TD. Common genetic determinants of vitamin D insufficiency: a genome-wide association study. *Lancet* 2010; 376: 180-188 [PMID: 20541252 DOI: 10.1016/S0140-6736(10)60588-0]

32. Ahn J, Yu K, Stolzenberg-Solomon R, Simon KC, Mc- Cullough ML, Gallicchio L, Jacobs EJ, Ascherio A, Helzlsouer K, Jacobs KB, Li Q, Weinstein SJ, Purdue M, Virtamo J, Horst R, Wheeler W, Chanock S, Hunter DJ, Hayes RB, Kraft P, Albanes D. Genome-wide association study of circulating vitamin D levels. *Hum Mol Genet* 2010; 19: 2739-2745 [PMID: 20418485 DOI: 10.1093/hmg/ddq155]

33. Powe CE, Evans MK, Wenger J, Zonderman AB, Berg AH, Nalls M, Tamez H, Zhang D, Bhan I, Karumanchi SA, Powe NR, Thadhani R. Vitamin D-binding protein and vitamin D status of black Americans and white Americans. *N Engl*

J Med 2013; 369: 1991-2000 [PMID: 24256378 DOI: 10.1056/ NEJMoa1306357]

34. Tanaka A, Nezu S, Uegaki S, Kikuchi K, Shibuya A, Miyakawa H, Takahashi S, Bianchi I, Zermiani P, Podda M, Ohira H, Invernizzi P, Takikawa H. Vitamin D receptor polymorphisms are associated with increased susceptibility to primary biliary cirrhosis in Japanese and Italian populations. *J Hepatol* 2009; 50: 1202-1209 [PMID: 19376604 DOI: 10.1016/j.jhep.2009.01.015]

35. Fan L, Tu X, Zhu Y, Zhou L, Pfeiffer T, Feltens R, Stoecker W, Zhong R. Genetic association of vitamin D receptor polymorphisms with autoimmune hepatitis and primary biliary cirrhosis in the Chinese. *J Gastroenterol Hepatol* 2005; 20: 249-255 [PMID: 15683428]

36. Vogel A, Strassburg CP, Manns MP. Genetic association of vitamin D receptor polymorphisms with primary biliary cirrhosis and autoimmune hepatitis. *Hepatology* 2002; 35: 126-131 [PMID: 11786968]

37. Halmos B, Szalay F, Cserniczky T, Nemesanszky E, Lakatos P, Barlage S, Schmitz G, Romics L, Csaszar A. Association of primary biliary cirrhosis with vitamin D receptor BsmI genotype polymorphism in a Hungarian population. *Dig Dis Sci*2000; 45: 1091-1095 [PMID: 10877221]

38. Baur K, Mertens JC, Schmitt J, Iwata R, Stieger B, Eloranta JJ,Frei P, Stickel F, Dill MT, Seifert B, Ferrari HA, von Eckardstein A, Bochud PY, Müllhaupt B, Geier A. Combined effect of 25-OH vitamin D plasma levels and genetic vitamin D receptor (NR 1I1) variants on fibrosis progression rate in HCV

patients. *Liver Int* 2012; 32: 635-643 [PMID: 22151003 DOI:10.1111/j.1478-3231.2011.02674]

39. Falleti E, Bitetto D, Fabris C, Cussigh A, Fontanini E, FornasiereE, Fumolo E, Bignulin S, Cmet S, Minisini R, Pirisi M, Toniutto P. Vitamin D receptor gene polymorphisms and hepatocellular carcinoma in alcoholic cirrhosis. *World J Gastroenterol* 2010; 16: 3016-3024 [PMID: 20572305]
40. The National Academies Collection: Reports funded byNational Institutes of Health; Ross AC, Taylor CL, Yaktine AL, Del Valle HB, editors. Institute of Medicine (US) Committee to Review Dietary Reference Intakes for Vitamin D and Calcium. The National Academies Collection: Reports unded by National Institutes of Health. Dietary Reference Intakes for Calcium and Vitamin D. Washington (DC): National Academies Press (US), 2011 [PMID: 21796828]
41. Holick MF, Binkley NC, Bischoff-Ferrari HA, Gordon CM, Hanley DA, Heaney RP, Murad MH, Weaver CM. Evaluation, treatment, and prevention of vitamin D deficiency: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab* 2011; 96: 1911-1930 [PMID: 21646368]
42. European Association for the Study of the Liver. EASL Clinical Practice Guidelines: management of cholestatic liver diseases. *J Hepatol* 2009; 51: 237-267 [PMID: 19501929 DOI: 10.1016/j.jhep.2009.04.009]
43. Abramovitch S, Dahan-Bachar L, Sharvit E, Weisman Y, Ben Tov A, Brazowski E, Reif S. Vitamin D inhibits proliferation and profibrotic marker expression in hepatic stellate cells and decreases thioacetamide-induced liver

fibrosis in rats. *Gut* 2011; 60: 1728-1737 [PMID: 21816960 DOI: 10.1136/gut.2010.234666]

44. Neeman R, Abramovitch S, Sharvit E, Elad-Sfadia G, Haklai R, Kloog Y, Reif S. Vitamin D and S-farnesylthiosalicylic acid have a synergistic effect on hepatic stellate cells proliferation. *Dig Dis Sci* 2014; 59: 2462-2469 [PMID: 24942325]

45. Murillo G, Nagpal V, Tiwari N, Benya RV, Mehta RG. Actions of vitamin D are mediated by the TLR4 pathway in inflammation-induced colon cancer. *J Steroid Biochem Mol Biol* 2010; 121: 403-407 [PMID: 20214986 DOI: 10.1016/j. jsbmb. 2010.03.009]

46. Dickie LJ, Church LD, Coulthard LR, Mathews RJ, Emery P, McDermott MF. Vitamin D3 down-regulates intracellular Toll-like receptor 9 expression and Toll-like receptor 9-induced IL-6 production in human monocytes. *Rheumatology* (Oxford) 2010; 49: 1466-1471 [PMID: 20435648]

47. Lemire JM, Archer DC, Beck L, Spiegelberg HL. Immunosuppressive actions of 1,25-dihydroxyvitamin D3: preferential inhibition of Th1 functions. *J Nutr* 1995; 125: 1704S-1708S [PMID: 7782931]

48. Penna G, Adorini L. 1 Alpha,25-dihydroxyvitamin D3 inhibits differentiation, maturation, activation, and survival of dendritic cells leading to impaired alloreactive T cell activation. *J Immunol* 2000; 164: 2405-2411 [PMID: 10679076]

49. Boonstra A, Barrat FJ, Crain C, Heath VL, Savelkoul HF, O' Garra A. 1alpha,25-Dihydroxyvitamin d3 has a direct effect on naive CD4(+) T cells to

enhance the development of Th2 cells. *J Immunol* 2001; 167: 4974-4980 [PMID: 11673504]

50. Shirakawa H, Matsumoto A, Joshita S, Komatsu M, Tanaka N, Umemura T, Ichijo T, Yoshizawa K, Kiyosawa K, Tanaka E. Pretreatment prediction of virological response to peginterferon plus ribavirin therapy in chronic hepatitis C patients using viral and host factors. *Hepatology* 2008; 48: 1753-1760 [PMID: 18925643 DOI: 10.1002/hep.22543]

51. Gal-Tanamy M, Bachmetov L, Ravid A, Koren R, Erman A,Tur-Kaspa R, Zemel R. Vitamin D: an innate antiviral agent suppressing hepatitis C virus in human hepatocytes. *Hepatology* 2011; 54: 1570-1579 [PMID: 21793032 DOI: 10.1002/hep.24575]

52. Bitetto D, Fattovich G, Fabris C, Ceriani E, Falleti E, Fornasiere E, Pasino M, Ieluzzi D, Cussigh A, Cmet S, Pirisi M, Toniutto P. Complementary role of vitamin D deficiency and the interleukin-28B rs12979860 C/T polymorphism in predicting antiviral response in chronic hepatitis C. *Hepatology* 2011; 53: 1118-1126 [PMID: 21480318 DOI: 10.1002/ hep.24201]

53. Kitson MT, Dore GJ, George J, Button P, McCaughan GW, Crawford DH, Sievert W, Weltman MD, Cheng WS, Roberts SK. Vitamin D status does not predict sustained virologic response or fibrosis stage in chronic hepatitis C genotype 1 infection. *J Hepatol* 2013; 58: 467-472 [PMID: 23183524 DOI: 10.1016/j.jhep.2012.11.017]

54. Lange CM, Bojunga J, Ramos-Lopez E, von Wagner M, Hassler A, Vermehren J, Herrmann E, Badenhoop K, Zeuzem S, Sarrazin C. Vitamin D deficiency

and a CYP27B1-1260 promoter polymorphism are associated with chronic hepatitis C and poor response to interferon-alfa based therapy. *J Hepatol* 2011; 54: 887-893 [PMID: 21145801 DOI: 10.1016/ j.jhep.2010.08.036]

55. Bailey R, Cooper JD, Zeitels L, Smyth DJ, Yang JH, Walker NM, Hyppönen E, Dunger DB, Ramos-Lopez E, Badenhoop K, Nejentsev S, Todd JA. Association of the vitamin D metabolism gene CYP27B1 with type 1 diabetes. *Diabetes* 2007; : 2616-2621 [PMID: 17606874]

56. Alvarez JA, Ashraf A. Role of vitamin d in insulin secretion and insulin sensitivity for glucose homeostasis. *Int J Endocrinol* 2010; 2010: 351385 [PMID: 20011094 DOI: 10.1155/ 2010/351385]

57. Borissova AM, Tankova T, Kirilov G, Dakovska L, Kovacheva R. The effect of vitamin D3 on insulin secretion and peripheral insulin sensitivity in type 2 diabetic patients. *Int J Clin Pract* 2003; 57: 258-261 [PMID: 12800453]

58. Resnick LM. Calcium metabolism in hypertension and allied metabolic disorders. *Diabetes Care* 1991; 14: 505-520 [PMID:1864222]

59. Farrell GC, Larter CZ. Nonalcoholic fatty liver disease: from steatosis to cirrhosis. *Hepatology* 2006; 43: S99-S112 [PMID:16447287]

60. Song Y, Wang L, Pittas AG, Del Gobbo LC, Zhang C, Manson JE, Hu FB. Blood 25-hydroxy vitamin D levels and incident type 2 diabetes: a meta-analysis of prospective studies. *Diabetes Care* 2013; 36: 1422-1428 [PMID: 23613602 DOI: 10.2337/dc12-0962]

61. Afzal S, Brøndum-Jacobsen P, Bojesen SE, Nordestgaard BG. Vitamin D concentration, obesity, and risk of diabetes: a mendelian randomisation study.

Lancet Diabetes Endocrinol 2014; 2: 298-306 [PMID: 24703048 DOI: 10.1016/S2213-8587(13)70200-6]

62. Ford ES, Ajani UA, McGuire LC, Liu S. Concentrations of serum vitamin D and the metabolic syndrome among U.S. adults. *Diabetes Care* 2005; 28: 1228-1230 [PMID: 15855599]

63. Stein EM, Strain G, Sinha N, Ortiz D, Pomp A, Dakin G, McMahon DJ, Bockman R, Silverberg SJ. Vitamin D insufficiency prior to bariatric surgery: risk factors and a pilot treatment study. *Clin Endocrinol* (Oxf) 2009; 71: 176-183 [PMID:19018785 DOI: 10.1111/j.1365-2265.2008.03470.x]

64. Liu E, Meigs JB, Pittas AG, McKeown NM, Economos CD, Booth SL, Jacques PF. Plasma 25-hydroxyvitamin d is associated with markers of the insulin resistant phenotype in nondiabetic adults. *J Nutr* 2009; 139: 329-334 [PMID: 19106328 DOI: 10.3945/jn.108.093831]

65. Hitman GA, Mannan N, McDermott MF, Aganna E, Ogunkolade BW, Hales CN, Boucher BJ. Vitamin D receptor gene polymorphisms influence insulin secretion in Bangladeshi Asians. *Diabetes* 1998; 47: 688-690 [PMID: 9568705]

66. Wortsman J, Matsuoka LY, Chen TC, Lu Z, Holick MF. Decreased bioavailability of vitamin D in obesity. *Am J Clin Nutr* 2000; 72: 690-693 [PMID: 10966885]

67. Yanoff LB, Parikh SJ, Spitalnik A, Denkinger B, Sebring NG, Slaughter P, McHugh T, Remaley AT, Yanovski JA. The prevalence of hypovitaminosis D and secondary hyperparathyroidism in obese Black Americans. *Clin Endocrinol* (Oxf)

68. Goldner WS, Stoner JA, Thompson J, Taylor K, Larson L, Erickson J, McBride C. Prevalence of vitamin D insufficiency and deficiency in morbidly obese patients: a comparison with non-obese controls. *Obes Surg* 2008; 18: 145-150 [PMID: 18175194 DOI: 10.1007/s11695-007-9315-8]

69. Barchetta I, Angelico F, Del Ben M, Baroni MG, Pozzilli P, Morini S, Cavallo MG. Strong association between non alcoholic fatty liver disease (NAFLD) and low 25(OH) vitaminD levels in an adult population with normal serum liver enzymes. *BMC Med* 2011; 9: 85 [PMID: 21749681 DOI: 10.1186/1741-7015-9-85]

70. Dasarathy J, Periyalwar P, Allampati S, Bhinder V, Hawkins C, Brandt P, Khiyami A, McCullough AJ, Dasarathy S. Hypovitaminosis D is associated with increased whole body fat mass and greater severity of non-alcoholic fatty liver disease. *Liver Int* 2014; 34: e118-e127 [PMID: 24118743 DOI: 10.1111/liv.12312]

71. Day CP. Non-alcoholic fatty liver disease: current concepts and management strategies. *Clin Med* 2006; 6: 19-25 [PMID: 16521351]

72. Targher G. Non-alcoholic fatty liver disease and cardiovascular disease risk. *Curr Cardiovasc Risk Rep* 2010; 4: 32-39

73. Tilg H, Moschen AR. Evolution of inflammation in non-alcoholic fatty liver disease: the multiple parallel hits hypothesis. *Hepatology* 2010; 52: 1836-1846 [PMID: 21038418 DOI:

74. Donato MT, Lahoz A, Jiménez N, Pérez G, Serralta A, Mir J, Castell JV, Gómez-Lechón MJ. Potential impact of steatosis on cytochrome P450 enzymes

of human hepatocytes isolated from fatty liver grafts. *Drug Metab Dispos* 2006; 34: 1556-1562 [PMID: 16763015]

75. Roth CL, Elfers CT, Figlewicz DP, Melhorn SJ, Morton GJ, Hoofnagle A, Yeh MM, Nelson JE, Kowdley KV. Vitamin D deficiency in obese rats exacerbates nonalcoholic fatty liver disease and increases hepatic resistin and Toll-like receptor activation. *Hepatology* 2012; 55: 1103-1111 [PMID: 21994008 DOI: 10.1002/hep.24737]

PROFORMA

PROFORMA

CASE NO-

- Name:
- Age/sex:
- Address:
- IPNO:

QUESTIONNAIRES

- Duration of illness-
- Co morbid conditions:
- DM/Renal failure/Ischemic heart disease/Malignancy/others..
- History of recent drug intake:
- History of native treatment :
- Alteration of sleep pattern and behavior:

CLINICAL EXAMINATION

- General examination-
- Signs of liver cell failure-

SYSTEMIC EXAMINATION

- Abdomen-
- CVS -

- RS -
- CNS

INVESTIGATIONS

- Hb
- TC
- DC-P/L/M
- ESR
- RBS
- S.creatinine
- S.Vit 25(OH)D
- Viral markers-HBsAg,Anti HCV
- Sr. sodium
- Sr. Potassium
- Sr.T.protein,sr.albumin &globulin
- Sr.bilirubin
- Sr.SGPT,SGOT
- PT,INR
- Ascitic fluid analysis(TC,DC)
- Ultrasonography of abdomen
- Upper GI Scopy

DIAGNOSIS-

DURATION OF HOSPITAL STAY-

OUTCOME-

PATIENT CONSENT FORM

சுயஒப்புதல் படிவம்

ஆய்வு செய்யப்படும் தலைப்பு:

இடம்: பொது மருத்துவத்துவ துரை

அரசு கீழ்பாக்கம் மருத்துவ கல்லூரி மருத்துவமனை

சென்னை

பங்குபெறுபவரின் பெயர் :

பங்குபெறுபவரின் வயது :

பங்குபெறுபவரின் எண் :

மேலே குறிப்பிட்டுள்ள மருத்துவ ஆய்வின் விவரங்கள் எனக்கு விளக்கப்பட்டது. நான் இவ்வாய்வில் தன்னிச்சையாக பங்கேற்கிறேன்.எந்த காரணத்தினாலோ எந்த சட்டசிக்கலுக்கும் உட்படாமல் நான் இவ்வாய்வில் இருந்து விலகிக்கொள்ளல்லாம் என்றும் அறிந்து கொண்டேன்.

இந்த ஆய்வு சம்பந்தமாகவோ, இதை சார்ந்து மேலும் ஆய்வு மேற்கொள்ளும்போதும் இந்த ஆய்வில்பங்கு பெறும் மருத்துவர் என்னுடைய மருத்துவ அறிக்கைகளை பார்ப்பதற்கு என் அனுமதி தேவையில்லை என அறிந்து கொள்கிறேன். இந்த ஆய்வின் மூலம் கிடைக்கும் தகவலையோ, முடிவையோ பயன்படுத்திக்கொள்ள மறுக்க மாட்டேன்.

இந்த ஆய்வில் பங்கு கொள்ள ஒப்புக்கொள்கிறேன். இந்த ஆய்வை மேற்கொள்ளும் மருத்துவ அணிக்கு உண்மையுடன் இருப்பேன் என்றும் உறுதியளிக்கிறேன்.

பங்கேற்பவரின் கையொப்பம்

ஆய்வாளரின் கையொப்பம்

இடம் :

தேதி :

MASTER CHART

S.NO	AGE	SEX	BMI	VIT-D LEVEL(ng/ml)		TS.BILIRUBIN (mg/dl)	S.ALBUMIN (g/dl)	S.CREATININE (mg/dl)	INR	PT(pro)	ASCITIES	HE	ALCOHOLIC	AUTO IMMUNE PROFILE	VIRAL MARKER	CRYPTOGENIC	CHILD PUGH SCORE	CLASS	MELD SCORE	% MORTALITY	RBS
1	35	M	18.6	32	o	2.7	3.8	1	0.8	3.5	NO	NO	NO	NO	Y	NO	6	A	10	6%	109
2	45	M	23.4	28	i	3.6	3.2	0.8	1.6	3.6	EC	NO	Y	NO	NO	NO	9	B	17	6%	104
3	44	M	20.3	28	i	4	3.2	1.1	1.4	3.4	EC	NO	Y	NO	NO	NO	9	B	16	6%	103
4	56	M	21.5	29	i	2.6	3.8	1	1.5	3.2	NO	NO	NO	NO	NO	Y	6	A	15	6%	99
5	49	M	20.5	27	i	4.6	3.5	0.7	1	3	EC	NO	Y	NO	NO	NO	9	B	12	6%	98
6	48	M	29	20	i	11.2	2.6	4.4	3	7	EC	ML	NO	NO	Y	NO	13	C	42	71.30%	89
7	30	M	16.5	30	i	3	3.6	0.4	1.3	2	NO	NO	Y	NO	NO	NO	6	A	14	6%	105
8	47	F	23.4	23	i	3.7	3.1	1.3	1.2	2.5	EC	NO	NO	NO	NO	Y	9	B	16	6%	102
9	46	M	24.1	34	i	2.9	3.7	0.5	0.9	2.6	NO	NO	Y	NO	NO	NO	6	A	10	6%	100
10	45	F	28.5	32	i	2.7	4.4	0.4	1.1	2.5	NO	NO	Y	NO	NO	NO	6	A	11	6%	89
11	49	M	27.2	19	d	9.4	2.1	6.1	5	7	EC	ML	Y	NO	NO	NO	13	C	50	71.30%	98
12	49	M	25.5	25	i	4	3.4	0.9	0.9	2.6	EC	NO	Y	NO	NO	NO	9	B	11	6%	98
13	48	M	23.6	15	d	8.6	2.7	4.5	6	7	EC	ML	Y	NO	NO	NO	13	C	49	71.30%	96
14	44	M	26.9	30	i	2	4.1	0.7	0.8	2	NO	NO	Y	NO	NO	NO	6	A	9	6%	94
15	63	M	23.1	18	d	8.2	2.8	4.5	1.4	5	EC	ML	NO	NO	NO	Y	12	C	32	52.60%	95
16	45	M	28.5	24	i	5	3.3	2	1.5	2	EC	NO	Y	NO	NO	NO	9	B	23	19.60%	97
17	46	M	31.5	19	d	12	2.6	7	4.5	7	EC	ML	Y	NO	NO	NO	13	C	51	71.30%	110
18	48	M	24.8	20	i	4	3.1	3	1.5	2.5	EC	NO	Y	NO	NO	NO	9	B	26	19.60%	106
19	48	M	24	16	d	11	2.6	4	2.3	6	EC	ML	NO	NO	Y	NO	12	C	38	52.60%	102
20	49	F	29.2	21	i	4.6	3.4	0.6	1.6	1.5	EC	NO	Y	NO	NO	NO	9	B	17	6%	95
21	47	M	24.6	21	i	5.8	3.3	0.8	1.2	1.8	EC	NO	Y	NO	NO	NO	9	B	15	6%	94
22	43	F	19.4	19	d	10.5	2.6	7.7	5	7	EC	ML	Y	NO	NO	NO	13	C	52	71.30%	93

23	34	M	17.6	35	o	2.5	3.9	0.7	1.4	1.8	NO	NO	Y	NO	NO	NO	6	A	13	6%	98
24	45	F	28	18	d	8.9	2.5	4	2.1	5	EC	ML	NO	NO	NO	Y	12	C	36	52.60%	104
25	48	M	21.2	25	i	6.7	3.5	0.7	1.5	1.5	EC	NO	Y	NO	NO	NO	9	B	18	6%	102
26	44	M	20.4	27	i	3.8	3	0.6	1.1	1.8	EC	NO	NO	NO	Y	NO	9	B	12	6%	94
27	45	M	20.5	22	i	5.6	3.3	0.9	1.2	2.5	EC	NO	Y	NO	NO	NO	9	B	15	6%	104
28	46	F	21.3	21	i	5	3.5	0.8	1.5	2	EC	NO	Y	NO	NO	NO	9	B	17	6%	89
29	59	F	25.7	33	o	2.9	3.9	0.8	1.3	3	NO	NO	Y	NO	NO	NO	6	A	12	6%	102
30	43	M	23.6	16	d	11.9	2	2.1	6.5	7	EC	ML	Y	NO	NO	NO	13	C	43	71.30%	88
31	49	M	31.5	20	i	4.7	3.2	0.7	1.6	3.5	EC	NO	Y	NO	NO	NO	9	B	17	6%	87
32	46	M	32	18	d	9	2.7	3	2.2	5	EC	ML	Y	NO	NO	NO	11	C	34	52.60%	86
33	44	F	21.1	28	i	7.5	3.1	0.7	1.2	2.5	EC	NO	Y	NO	NO	NO	9	C	16	6%	105
34	44	M	24.1	27	i	6.8	3.5	0.7	1.3	3	EC	NO	Y	NO	NO	NO	9	C	16	6%	103
35	47	M	23.5	35	o	2.8	4.1	0.7	1.2	2.5	NO	NO	Y	NO	NO	NO	6	A	12	6%	103
36	20	F	21.2	36	o	2.9	4.2	0.7	1.3	2.5	NO	NO	Y	Y	NO	NO	6	A	13	6%	102
37	48	M	22.2	17	d	8.6	2.6	2.2	1.9	6	EC	ML	Y	NO	NO	NO	12	C	29	19.60%	101
38	47	M	28.8	27	i	3.9	3.5	0.7	1.1	3	EC	NO	Y	NO	NO	NO	9	B	12	6%	100
39	46	M	23.1	14	d	9.5	2.3	1.5	1.1	4	EC	ML	Y	NO	NO	NO	12	C	19	6%	98
40	46	F	19.5	14	d	10.2	2.1	1.6	2.3	6	EC	ML	Y	NO	NO	NO	12	C	29	19.60%	95
41	48	M	20.2	26	i	6.5	3.4	0.8	1.2	3	EC	NO	Y	NO	NO	NO	9	B	15	6%	94
42	43	M	18.5	22	i	5.8	3.3	0.8	0.9	3	EC	NO	Y	NO	NO	NO	9	B	13	6%	92
43	46	M	21.4	13	d	11.5	2.9	0.8	2	5	EC	ML	Y	NO	NO	NO	12	C	23	19.60%	100
44	60	M	21.7	25	i	6	3.2	0.8	1.1	1.5	EC	NO	NO	NO	Y	NO	9	B	14	6%	103
45	47	M	24.5	23	i	5	3.1	0.9	1.1	1.6	EC	NO	Y	NO	NO	NO	9	B	13	6%	103
46	45	M	23.3	24	i	3.9	3	1	1.5	1.5	EC	NO	NO	NO	NO	Y	9	B	16	6%	104
47	44	F	30	20	i	11	2.6	2.2	1.8	6	EC	ML	Y	NO	NO	NO	12	C	29	19.60%	90
48	48	M	19.9	32	o	2.5	4	0.8	1.4	2.5	NO	NO	NO	NO	NO	Y	6	A	13	6%	84
49	43	M	25.4	23	i	5	3.5	1.2	1.5	2.8	EC	NO	Y	NO	NO	NO	9	B	18	6%	96
50	46	M	27	31	o	2.4	3.9	0.9	1.2	3	NO	NO	NO	NO	Y	NO	6	A	11	6%	102
51	34	M	18.2	22	i	4.5	3.4	0.9	1.6	3	EC	NO	Y	NO	NO	NO	9	B	17	6%	102

52	45	M	21.6	10	d	8	2.5	2.5	2.2	5	EC	ML	Y	NO	NO	NO	12	C	31	52.60%	106
53	47	M	20.5	20	i	4	3.1	0.8	1.6	3.8	EC	NO	Y	NO	NO	NO	9	B	16	6%	98
54	45	F	26	30	i	2.2	3.6	0.7	1.2	2.5	NO	NO	Y	NO	NO	NO	6	A	11	6%	99
55	44	M	18.2	19	d	3.5	3.5	0.8	1.3	2	EC	NO	Y	NO	NO	NO	8	C	14	6%	94

KEY TO MASTER CHART

ML	-	Minimal
EC	-	Easily controlled
i	-	insufficient
d	-	deficient
O	-	Optimal
Y	-	Yes